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**US Army Corps
of Engineers**



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**AQUATIC PLANT CONTROL
RESEARCH PROGRAM**

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**PROCEEDINGS,
26TH ANNUAL MEETING,
AQUATIC PLANT CONTROL
RESEARCH PROGRAM**

**18-22 NOVEMBER 1991
DALLAS, TEXAS**

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June 1992

Final Report

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<p>The 26th Annual Meeting of the US Army Corps of Engineers Aquatic Plant Control Research Program was held in Dallas, TX, on 18-22 November 1991, to review current research activities and to afford an opportunity for presentation of operational needs. Papers presented at the meeting are included in this report.</p>			
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Preface

The 26th Annual Meeting of the US Army Corps of Engineers Aquatic Plant Control Research Program (APCRP) was held in Dallas, TX, on 18-22 November 1991. The meeting is required by Engineer Regulation 1130-2-412, paragraph 4c, and was organized by personnel of the APCRP, which is managed under the Environmental Resources Research and Assistance Programs (ERRAP) of the Environmental Laboratory (EL), US Army Engineer Waterways Experiment Station (WES), Vicksburg, MS.

The organizational activities were carried out and presentations by WES personnel were prepared under the general supervision of Mr. J. L. Decell, Program Manager,

ERRAP, EL. Mr. Robert C. Gunkel, Assistant Program Manager, ERRAP, was responsible for planning the meeting. Dr. John Harrison was Director, EL, WES. Mr. James W. Wolcott was Technical Monitor for the Headquarters, US Army Corps of Engineers.

The report was edited by Ms. Jessica S. Ruff of the Information Technology Laboratory (ITL), WES. Ms. Betty Watson, ITL, designed and composed the layout.

At the time of publication of this report, Director of WES was Dr. Robert W. Whalin. Commander and Deputy Director was COL Leonard G. Hassell, EN.

Agenda

Monday, 18 November 1991

- | | |
|------------|--|
| 1:00 p.m. | <i>Registration</i> |
| -5:00 p.m. | (Ballroom Foyer) |
| 1:00 p.m. | <i>Texas Aquatic Plant Management Society Meeting</i> |
| -6:00 p.m. | (Lincoln Room—1, 2, and 3) |
| 6:00 p.m. | <i>Reception</i> |
| -7:30 p.m. | (Lobby Court) |

Tuesday, 19 November 1991

- | | |
|------------|---|
| 8:00 a.m. | <i>Registration</i> |
| -5:00 p.m. | (Ballroom Foyer) |
| 8:00 a.m. | <i>Poster and Demonstration Session</i> |
| -5:00 p.m. | (Jackson Room) |
| 8:30 a.m. | <i>General Session</i> |
| -1:45 p.m. | (Lincoln Room—East, 1, 2, and 3) |
| 8:30 a.m. | Call to Order and Announcements
* Robert C. Gunkel, Assistant Manager, Aquatic Plant Control
Research Program (APCRP)
Waterways Experiment Station (WES)
Vicksburg, Mississippi |
| 8:40 a.m. | Welcome to Fort Worth District
* COL John A. Mills, Commander
U.S. Army Engineer (USAE) District
Fort Worth, Texas |
| 8:50 a.m. | Comments by the Program Manager
* J. Lewis Decell, WES |
| 9:00 a.m. | Wetland Issues and Possible Impacts on Aquatic Plant Control Activities
* Russell F. Theriot, WES |

- 9:30 am. .Aquatic Plant Control Operations Support Center (APCOSC) Update
* William C. Zattau, USAE District
Jacksonville, Florida
- 9:45 a.m. Lewisville Aquatic Ecosystem Research Facility (LAERF) Update (32733)
* R. Michael Smart, WES, LAERF
Lewisville, Texas
- 10:05 a.m. ***Break***
- 10:35 a.m. Economics and Aquatic Plant Management (32729)
* Jim E. Henderson, WES
- 10:50 a.m. Movement and Habitat Utilization of Triploid Grass Carp (32738)
* Jeffrey W. Foltz, Clemson University
Clemson, South Carolina

Simulation Technology

R. Michael Stewart, WES, Presiding

- 11:05 a.m. Role of Simulation Technology in the APCRP
* R. Michael Stewart, WES
- 11:25 a.m. Status and Application of the WES AMUR/STOCK and INSECT Models (32438)
* William A. Boyd, WES
- 11:45 a.m. Status and Application of the WES HERBICIDE Model (32439)
* Philip A. Clifford, EA Engineering, Science, and Technology, Inc.
Sparks, Maryland
- 12:00 noon ***Lunch***
- 1:00 p.m. Updates to Plant Growth Models for Hydrilla and Milfoil (32440)
* R. Michael Stewart, WES
- 1:15 p.m. Studies for Further Development of Existing WES Plant Growth Models (32440)
* John D. Madsen, WES, LAERF
- 1:30 p.m. Utilization of GIS/DBMS Techniques to Support User Application of APC
Simulation Models (32506)
* M. Rose Kress, WES
- 1:45 p.m. ***Adjourn General Session***
- 2:00 p.m. ***USAЕ Division/District Working Session***
-5:00 p.m. (Washington and Adams (A) Rooms)

Wednesday, 20 November 1991

- 7:00 a.m. **Federal Aquatic Plant Management Working Group**
-9:00 a.m. (Audubon Room)

Agenda

8:00 a.m. ***Poster and Demonstration Session***
-12:30 p.m. Jackson Room

9:00 a.m. ***General Session***
-12:30 p.m. (Lincoln Room—East, 1, 2, and 3)

Chemical Technology

Kurt D. Getsinger, WES, Presiding

- 9:00 a.m. Chemical Control Technology: Overview
* Kurt D. Getsinger, WES
- 9:15 a.m. Herbicide Concentration/Exposure Time Relationships (32352)
* Michael D. Netherland, WES
- 9:30 a.m. Herbicide Flume Studies at TVA-ARL
* E. Glenn Turner, WES
- 9:45 a.m. Herbicide Delivery Systems (32437)
* Michael D. Netherland, WES
- 10:00 a.m. Field Evaluations of a Slow-Release Matrix Device in Flowing Water
* David Sisneros, U.S. Bureau of Reclamation
Denver, Colorado
- 10:15 a.m. Submersed Application Techniques for Flowing Water (32354)
* Kurt D. Getsinger, WES
- 10:30 a.m. ***Break***
- 11:00 a.m. Field Evaluation of Selected Herbicides (32404)
* Kurt D. Getsinger, WES
- 11:15 a.m. Plant Growth Regulators for Aquatic Plant Management (32578)
* Linda S. Nelson, WES
- 11:30 a.m. Evaluation of Mariner as a PGR on Hydrilla
* Thai K. Van, U.S. Department of Agriculture (USDA)
Fort Lauderdale, Florida
- 11:45 a.m. Potential PGRs for Aquatic Plant Management (32578)
* Carole A. Lembi, Purdue University
West Lafayette, Indiana
- 12:00 noon Phenology of Aquatic Plants (32441)
* John D. Madsen, WES, LAERF
- 12:15 p.m. Relationship Between Plant Hormones and Carbohydrate Partitioning in
Monoecious Hydrilla (32441)
* Stephen J. Klaine, Memphis State University
Memphis, Tennessee
- 12:30 p.m. ***Lunch***
- 2:00 p.m. Field Trip to Lewisville Aquatic Ecosystem Research Facility

- 5:30 p.m. Texas Barbecue Dinner
 7:30 p.m. Return to Doubletree Hotel

Thursday, 21 November 1991

- 8:00 a.m. ***General Session***
 -3:00 p.m. (Lincoln Room—East, 1, 2, and 3)

Ecological Technology

John W. Barko, WES, Presiding

- 8:00 a.m. An Overview of Ecological Studies
 * John W. Barko, WES
- 8:15 a.m. Effects of Sediment N Supply on Interactions Between *P. americanus* and *H. verticillata* (32351)
 * Nancy J. McCreary, Lafayette College
 Easton, Pennsylvania
- 8:30 a.m. Rooting Depth of *M. spicatum* in Relation to Sediment Fertility (32351)
 * Dwilette G. McFarland, WES
- 8:45 a.m. Aquatic Plant Competition Studies in Guntersville Reservoir (32736)
 * R. Michael Smart, WES, LAERF
- 9:00 a.m. Competitive Interactions Among Introduced and Native Species (32577)
 * R. Michael Smart, WES, LAERF
- 9:15 a.m. Submersed Macrophyte Invasions and Declines (32351 & 32405)
 * Craig S. Smith, WES
- 9:30 a.m. Habitat Value of Aquatic Macrophytes: Studies in Lake Seminole, Florida
 (32505)
 * Andrew C. Miller, WES
- 9:45 a.m. ***Break***
- 10:15 a.m. Comparison of Largemouth Bass Growth Between *Hydrilla verticillata* and *Potamogeton nodosus* Macrophyte Beds (32505)
 * K. Jack Killgore, WES
- 10:30 a.m. Seasonal Variations in Nighttime Convective Circulation and Phosphorus Transport Between Macrophyte Beds and Open Water (32405)
 * William F. James, WES, Eau Gallie Laboratory
 Spring Valley, Wisconsin
- 10:45 a.m. Effects of Benthic Barrier Placement on Aquatic Habitat Conditions (32579 & 32737)
 * Harry L. Eakin, WES

Agenda

Biological Technology

Alfred F. Cofrancesco, WES, Presiding

- 11:00 a.m. Overview on Biological Control
* Alfred F. Cofrancesco, WES
- 11:15 a.m. Insect Biocontrol Agents of Hydrilla in Florida (31799)
* Ted D. Center, USDA
Fort Lauderdale, Florida
- 11:30 a.m. Temperate Biocontrol Insects for Eurasian Watermilfoil and Hydrilla (32730)
* Gary R. Buckingham, USDA
Gainesville, Florida
- 11:45 a.m. Release and Establishment of *Hydrellia* Flies (31799 & 32734)
* Michael J. Grodowitz, WES
- 12:00 noon **Lunch**
- 1:00 p.m. The Impact of Temperature on *Hydrellia pakistanae* (32734)
* Ramona H. Warren, WES
- 1:15 p.m. Biological Control of Pistia (32406)
* F. Allen Dray, USDA
Fort Lauderdale, Florida
- 1:30 p.m. Insect Biocontrol Agent of Eurasian Watermilfoil (32739)
* Sallie P. Sheldon, Middlebury College
Middlebury, Vermont
- 1:45 p.m. Biological Control Studies of Eurasian Watermilfoil Using Plant
Pathogens (32202)
* James P. Stack, ECOSCIENCE
Amherst, Massachusetts
- 2:00 p.m. Pathogen Biocontrol Studies of Hydrilla and Eurasian Watermilfoil
(32200 & 32735)
* Judy F. Shearer, WES
- 2:15 p.m. Biological Management of Aquatic Plants with Allelopathic and
Competitive Species (32408)
* Harvey L. Jones, WES
- 2:30 p.m. Report on Tuesday's Division/District Working Session
* William C. Zattau, USAE District
Jacksonville, Florida
- 3:00 p.m. ADOURN 26th Annual Meeting
- 3:00 **FY93 CIVIL WORKS R&D PROGRAM REVIEW**
- 5:00 p.m. (Corps of Engineers Representatives Only)
(Lincoln Room—1, 2, and 3)

FRIDAY, 22 NOVEMBER 1991

8:00 -12:00 noon **Joint Agency Guntersville Project - Principal Investigators Meeting**
(Room To Be Announced)

Posters and Demonstrations

Poster Presentations

Effects of Wetting and Drying Cycles on Vegetation Management in Ponds (32733)
* Gary O. Dick and R. Michael Smart, WES, LAERF

Effects of Aquatic Plants on Water Quality in Pond Ecosystems (32733)
* David R. Honnell, John D. Madsen, and R. Michael Smart, WES, LAERF

Environmental Characteristics of Ponds at the Lewisville Aquatic Ecosystem
Research Facility (32733)
* R. Michael Smart, WES, LAERF

Integration of GPS/GIS Technologies for Aquatic Plant Management
* R. Michael Stewart, WES

Knowledge-Based Systems for Aquatic Plant Management
* R. Michael Stewart, WES

Production of Propagules of Native Aquatic Plant Species (32577)
* Susan E. Monteleone and R. Michael Smart, WES, LAERF

Seed Germination and Seedling Survival of Waterhyacinth
* Rebecca S. Westover and John D. Madsen, WES, LAERF

Sampling Fish in Submersed Aquatic Plants (32505)
* K. Jack Killgore, WES

Computer Demonstrations

HERBICIDE and INSECT Models

* William A. Boyd, WES, Philip A. Clifford, EA Engineering, Science, and
Technology, Inc., and R. Michael Stewart, WES

GIS and HARVEST, GIS and HYDRILLA, and GIS and MILFOIL
* R. Michael Stewart and E. May Causey, WES

Computer Display of a *Hydrellia* Fly Expert System

* Craig S. Smith, Michael J. Grodowitz, William A. Johnson, WES, and Richard L.
Deonier, University of Miami, Oxford, Ohio

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Conversion Factors, Non-SI to SI Units of Measurement

Non-SI units of measurement used in this report can be converted to SI units as follows:

Multiply	By	To Obtain
acres	4,046.873	square meters
acre-feet	1,233.489	cubic meters
feet	0.3048	meters
gallons (US liquid)	3.785412	liters
inches	2.54	centimeters
miles (US statute)	1.609347	kilometers
ounces (mass)	28.34952	grams
pounds (mass)	0.4535924	kilograms
tons (mass) per acre	0.22	kilograms per square meter

26th Annual Meeting US Army Corps of Engineers

AQUATIC PLANT CONTROL RESEARCH PROGRAM

Introduction

The Corps of Engineers (CE) Aquatic Plant Control Research Program (APCRP) requires that a meeting be held each year to provide for professional presentation of current research projects and to review current operations activities and problems. Subsequent to these presentations, the Civil Works Research and Development Program Review is held. This program review is attended by representatives of the Civil Works and Research Development Directorates of the Headquarters, US Army Corps of Engineers; the Program Manager, Environmental Resources Research and Assistance Programs (ERRAP); and representatives of the operations elements of various CE Division and District Offices.

The overall objective of this annual meeting is to thoroughly review the Corps aquatic plant control needs and establish priorities for future research, such that identified needs are satisfied in a timely manner.

The technical findings of each research effort conducted under the APCRP are reported to the Manager, ERRAP, US Army Engineer Waterways Experiment Station, each year in

the form of periodic progress reports and a final technical report. Each technical report is distributed widely in order to transfer technology to the technical community. Technology transfer to the field operations elements is effected through the conduct of demonstration projects in various District Office problem areas and through publication of Instruction Reports, Engineer Circulars, and Engineer Manuals. Periodically, results are presented through publication of an APCRP Information Exchange Bulletin which is distributed to both the field units and the general community. Public-oriented brochures, movies, and speaking engagements are used to keep the general public informed.

The printed proceedings of the annual meetings are intended to provide all levels of Corps management with an annual summary to ensure that the research is being focused on the current nationwide operational needs.

The contents of this report include the presentations of the 26th Annual Meeting held in Dallas, TX, on 18-22 November 1991.

Welcoming Remarks

by

COL John A. Mills¹

Welcome to Dallas and the Fort Worth District. We are honored that the Waterways Experiment Station has selected our District for the 26th annual Aquatic Plant Control Research Program meeting.

Many of you may wonder what type of aquatic plant problems we could possibly have in Texas! In the Fort Worth District, three of our 24 reservoirs constitute the bulk of the aquatic plant control operations. These reservoirs in east Texas have been periodically treated to rid them of aquatic growth where navigation and recreation activities are adversely impacted. Such controls are conducted with Corps Operations & Maintenance funds. Our neighbor to the south, the Galveston District, conducts most of the aquatic plant control and manages the Corps' Aquatic Plant Control Program in Texas. The Galveston District currently has a cooperative agreement with the Texas Parks and Wildlife Department, which performs the actual control efforts in Texas under the Aquatic Plant Control Program.

While control problems in Texas are not of the magnitude of those in other southern states, we do have our share of nuisance aquatic plants, most of which are courtesy of our neighbors to the east. Species of major concern in Texas are waterhyacinth, hydrilla, alligatorweed, and most recently, Brazilian elodea.

We cannot remain totally optimistic with regard to the aquatic plant situation within the State of Texas. Over the last several years we have seen the aquatic plant hydrilla gain a very significant foothold in our east Texas reservoirs. Let's not fool anybody; it is conceivable that this plant will spread to

some of our mainstream reservoirs on the Trinity and Brazos River systems. We have, at this time, a major infestation of hydrilla and Brazilian elodea at our Town Bluff Dam-B. A. Steinhagen Lake in the Piney Woods area of east Texas. We may well need the help of WES and the Texas Parks and Wildlife Department to combat this infestation. This particular lake is too small, too shallow, and too vulnerable to extensive hydrilla infestations on a year-to-year basis. Lake O'the Pines and Sam Rayburn have hydrilla infestations, though not as critical as the situation at Town Bluff Dam-B. A. Steinhagen Lake.

Aquatic plant control in Texas in the near future will be no easy ride. Money and manpower commitments will be needed. In a time of overall limited budgeting within the Corps, there will be no easy solution. The question will be, how critical is aquatic plant control—compared to other more routine and equally necessary O&M items.

I am encouraged that the Aquatic Plant Control Program continues to rise to the challenge of finding environmentally sound solutions to managing nuisance aquatic plant growth while ensuring that the multipurpose uses of our projects are maintained.

Most of you will be visiting the aquatic plant control research laboratory located at Lewisville Lake, a few miles north of here. The facility is a fine example of how innovative solutions to program needs can be found. In 1986, what is now the research facility was an abandoned state fish hatchery in dire need of maintenance and repair. When the Texas Parks and Wildlife Department decided to sell the property, the Fort Worth District purchased it. Happily, Mr. Lewis Decell, of

¹ Commander, US Army Engineer District, Fort Worth; Fort Worth, TX.

the Aquatic Plant Control Research Program, identified a need for a "real world" laboratory, and the facility at Lewisville received a new lease on life.

Following the 1988 memorandum of agreement between the Fort Worth District and WES, renovation of the 12.5-acre facility with 55 earthen ponds began. Dr. Michael Smart will be providing you with an update on the facility and current research efforts in a few minutes. We in the Fort Worth District

are very proud to have played a role in securing such a fine research facility.

Let me extend to you my wish for a productive and educational meeting. I understand that an old-fashioned Texas barbecue is planned for Wednesday evening at the Lewisville project. Those of you who attended the 1989 meeting in Huntsville, Alabama, will be pleased to know that we have checked the weather forecast, and tornadoes are NOT predicted. So, "you all" relax and enjoy our Texas hospitality.

Lewisville Aquatic Ecosystem Research Facility: An Update

by
*R. Michael Smart*¹

Introduction

The Waterways Experiment Station (WES) is operating an experimental pond facility located in Lewisville, Texas. The Lewisville Aquatic Ecosystem Research Facility (LAERF) is being developed under the auspices of the Aquatic Plant Control Research Program for studies of the biology, ecology, and control of aquatic plants. The LAERF receives partial funding directly from the APCRP, with the remainder of the funds provided by research projects of the resident staff and from fees charged to users of the ponds.

The objective of this article is to provide an update on the renovation, development, and operation of this unique facility. Topics to be covered include personnel, facilities, research, and future plans.

Personnel

An analytical laboratory manager position was added to the resident staff. The responsibilities of this new position include the development and operation of an onsite analytical laboratory to support research conducted at the LAERF. With the addition of this position, the resident staff now includes four employees: a pond/facilities manager, a lab manager, and two aquatic plant scientists. The facility also employs 10 to 12 part-time, graduate, and undergraduate contract students.

Facilities

To properly conduct research on the biology, ecology, and control of aquatic plants,

an onsite analytical capability was required. During the past year we have concentrated on acquiring this capability. We have recently obtained analytical equipment to process and analyze water, sediment, and plant tissue samples. Our analytical facilities are available to support APCRP research conducted at the LAERF. We have also initiated a water quality monitoring program to obtain basic information on pond water quality on a regular basis. This information is available to researchers using the ponds.

Although the LAERF is known primarily for the 55 earthen ponds that are available for experimental research on aquatic and wetland ecosystems, we also have been developing several other research facilities. These include a greenhouse tank facility and two flowing water raceway facilities. The greenhouse facility includes fifteen 1,200-liter fiberglass tanks equipped with individual temperature control units. These tanks can be filled with artificial or natural lake water, and are being used for conducting short-term, controlled studies to supplement longer term studies conducted in the ponds. One of the raceway facilities has been covered with a greenhouse and is being used for maintaining populations of waterhyacinths during winter periods. Both raceway facilities are being used for holding/culturing aquatic plants and for conducting research under flowing water conditions.

We are continuing to renovate the ponds based on demand and availability of funds. During fiscal year (FY) 1991, 5 of the 55 ponds were used for culture/study and 15 were used for research. Eight additional

¹ US Army Engineer Waterways Experiment Station, Lewisville Aquatic Ecosystem Research Facility, Lewisville, TX.

ponds have been reserved for FY 92, for a total anticipated use of 28 ponds. Three ponds require valve replacements, and 24 ponds are currently available for use. Excess ponds will be made available to researchers at other Federal and state agencies and universities.

Research

The LAERF is being used for research in support of each of the technology areas of the APCRP. Biological control is represented by studies of the application efficiency of different formulations of the microbial pathogen *Mycroleptodiscus terrestris*. Applications technology is being advanced in studies of the phenology or life cycle of waterhyacinth and in studies of the environmental effects of benthic barriers for control of submersed aquatic plants. Simulation technology is benefiting from the collection of additional data on biomass production and measurements of critical growth processes such as photosynthesis and respiration. Within the ecology area, studies are being conducted to understand fish-plant interactions and to determine the effects of weedy aquatic plant species on important fish such as largemouth bass. Other studies are examining the feasibility of replacing weedy species of aquatic plants with native, nonweedy species in order to provide beneficial habitat without the problems that excessive growth of weedy species can bring. Chemical control studies are currently in the planning stages (see next section, Future Plans).

In addition to the direct-allotted research, several reimbursable efforts are currently being conducted at LAERF. One project involves a greenhouse bioassay of the potential of sediments collected from Onondaga Lake, New York, to support the growth of aquatic plants. This work, funded by the US Environmental Protection Agency as a Clean Lakes Program Demonstration site, is concerned with identifying methods for reestablishing aquatic and wetland vegetation as an ameliorative treatment in this chronically polluted lake. LAERF personnel are also involved in field efforts such as an ecological assessment

of Kirk Pond, being conducted for the Portland District, Corps of Engineers, and a cooperative effort with the Tennessee Valley Authority aimed at establishing competitive species in Guntersville Reservoir in Alabama.

Future Plans

Renovation and development of the LAERF will continue during FY 92. The shed roof over one of the raceway facilities will be removed to make it more suitable for conducting aquatic plant research. Because of the high demand, five additional temperature-controlled fiberglass tanks will be added to the greenhouse facility. Additional laboratory space will be acquired to meet an increasing demand for laboratory facilities and to alleviate overcrowding.

A major addition to the research capabilities of the LAERF will be a large, outdoor mesocosm facility. This facility will include a water supply pond, 22 large fiberglass tanks, an effluent collection pond, a sediment preparation area, and a small laboratory. The mesocosm facility will be used primarily for conducting studies of the efficacy of aquatic herbicides and plant growth regulators (see **Chemical Control Technology** section). Research conducted in this facility will supplement laboratory/growth chamber studies conducted at the WES. The extension of this research to large-scale, outdoor systems will allow consideration of the effects of seasonal cycles and phenological stage of development on the effects of herbicides on both target and nontarget aquatic plants.

Acknowledgments

Many people have contributed to the continuing development of the LAERF. The contributions of the following, in particular, are gratefully acknowledged: J. T. Alewine, Rex Boyd, Michael Crouch, Gary Dick, Kurt Getsinger, Kay Glassie, Tammy Hancock, David Holland, David Honnell, John Madsen, Allen Martin, Kimberly Mauermann, Gracie Pfeiffer, Larry Prestien, Joe Snow, Mike Stewart, and Bekah Westover.

Economics and Aquatic Plant Control

by
Jim E. Henderson¹

Introduction

The invasion of waterways of the United States by exotic or nonnative aquatic vegetation has resulted in problems and impediments to the operation and public use of the waterways. The economic costs and benefits of plant control to the public and private sector are only beginning to be addressed. Public agency decisions on control technologies and long-term control strategies will benefit from information on issues of economic concern such as public perceptions of desirable plant densities and control technologies. Some limited information is available on these concerns (Milon, Yingling, and Reynolds 1986), and investigations are ongoing at Lake Guntersville, Alabama.

The purpose of this presentation is to examine the relation of aquatic plant infestations and control efforts to public economic benefits, and to provide a framework for the information being produced by the Guntersville study and other agency work. The primary emphasis here is on the effect of aquatic plants and aquatic plant control on nonmarket values associated with water resource projects. A more detailed examination of the impact of aquatic plants on market values is provided in Henderson (1991).

Benefits of Water Resource Development-Waterways

Public waterways are operated to provide a range of public goods and services, ranging from commercial navigation and flood control benefits, to outdoor recreation, wildlife, and scenic values. The economic benefits to individuals attributable to these goods and

services may be estimated through surveys or direct questioning, analysis of market data, or by judgments, e.g. voting, on alternatives (Freeman 1979).

Aquatic plant infestations result in changes in the economic benefits that result from these goods and services, by altering the ability of the natural resource to produce the good or by diminishing the quantity or quality and value of the good or service provided (Figure 1). A closer examination of these concepts is provided in the following paragraphs.

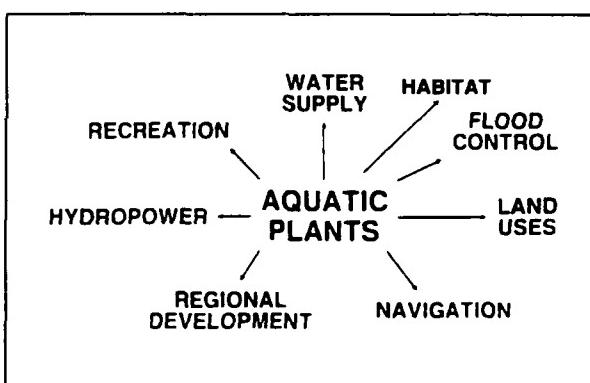


Figure 1. Changes in economic benefits as a result of aquatic plant infestations

Consumption of Public Goods and Services

Public waterways are operated as public resources (sometimes referred to as common-property resources) and produce public goods and services in the form of amenities as outdoor recreation, scenic values, and fishery habitat (Dorfman and Dorfman 1977). Several characteristics of public goods and services affect their consumption and valuation by the

¹ US Army Engineer Waterways Experiment Station, Vicksburg, MS.

public and influence decisions that are made about the resources.

The first characteristic is the nonexclusive nature of environmental resources. Environmental resources such as the air and water are not privately owned, and their use by other individuals cannot be excluded or prevented. The other important characteristic of public goods is the concept of externalities—those things that are important for an individual's value but are outside of his control. Externalities exist because, for instance, to benefit from a public good such as recreation, consumption by individuals requires not only certain components that they personally control (e.g., their time and expenditures for the activity) but also components that are outside the individual's control (things such as quality of the natural resource, availability of facilities, and scenic amenities).

Environmental amenities such as clean air, clean water, and access to recreation opportunities are the result of decisions made to ensure their provision. There are no property rights to public goods, and no compensation is required for their loss (Mishan 1969, as cited in Dorfman and Dorfman 1977). A variety of political and legal institutions and arrangements have been established to manage publicly held resources (Dales 1968, as cited in Dorfman and Dorfman 1977).

One aspect of joint consumption of publicly held resources is that use by one individual or group may prevent the use or diminish the quality of use by another individual or group. Because of the nonexclusive nature, public goods are subject to overuse through free and competitive use. Efforts to manage the productivity of the resource through regulation of consumption or use may be successful, but it is often difficult to separate the changes that result from a natural change in the resources from those that are attributable to management efforts (Gordon 1954, as cited in Dorfman and Dorfman 1977).

Demand for Public Goods

Total utility concepts

Provision of public goods and the consumption of those goods by the public are determined by availability of the public goods and the public demand for those goods and services. A generalized illustration of demand for a public good is shown in Figure 2. The y-axis represents "Total Utility" but can be thought of as a measure of value as Total Societal Good, Monetary Value, or some other measure of welfare. The x-axis shows "Quantity of X Consumed" with X being an environmental good such as recreation days, water quality, or, in this case, aquatic plant control.

This generalized graph shows the relation of providing public goods to the value those goods provide, starting from a zero level of the public good and increasing the quantity of the good consumed.

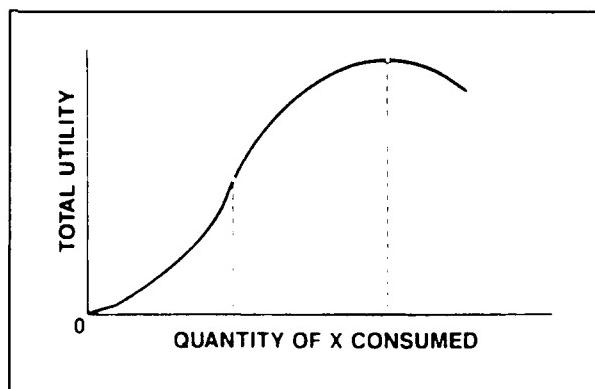


Figure 2. Relation of consumption of public goods to total utility

Starting at the zero level and increasing the quantity, it is observed that value rises rapidly with each marginal increase of the good.

This is consistent with conditions of high demand for the good in which little of the good is available. Where no aquatic plant

control is available, conditions may be such that boaters are unable to launch their boats or plant densities are so heavy that boat-houses and slips are completely blocked. Given these conditions, a minimal quantity of control will be highly valued, and will provide a high level of utility.

Examining the intermediate part of the graph, this shows that increasing the control still produces an increase in utility and value, but the rate of increase becomes more constant. This is consistent with an intermediate level of a service; the areas and users that produce the highest public value have been accommodated, and further increases in control still produce higher overall value.

The nature of aquatic plant control makes the development of demand or valuation curves somewhat more problematic than the generalized example in Figure 2. Long-term control strategies are often developed with some overall percent coverage of a lake as the long-range objective. This long-range approach is necessary given the limited funding each year and the year-to-year fluctuations in plant biomass, which are due to the natural resource changes that cause changes in plant growing conditions, e.g. floods or droughts. A demand curve for this situation could be as shown in Figure 3, showing the relation of total utility to percent coverage.

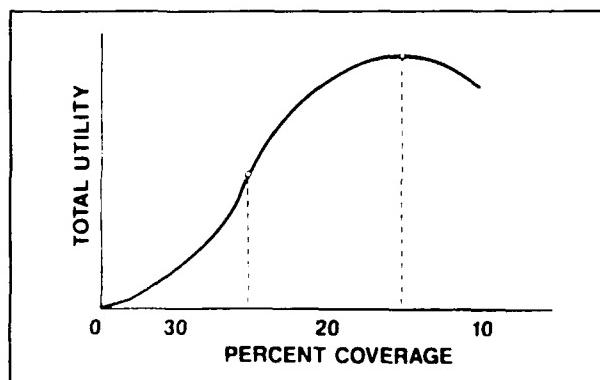


Figure 3. Relation of percentage aquatic plant coverage to total utility

Figures 2 and 3 give the impression of a continuous flow of plant control capacity, where the control is divisible and can be

stopped at any chosen point. Practically, to achieve a percentage control for an entire water body requires planning on the distribution of the control. The percentage of total control, along with criteria for control areas, is used to determine where and how often areas are treated. Because of the primacy of the specific coverage objectives (alternatives), the demand curve may be represented as a series of points representative of the alternatives, as shown in Figure 4.

Valuation work being planned for Lake Guntersville (Contingent Valuation Method) will present three alternative levels of control for recreation users to respond to regarding potential changes in their recreation use and their value for recreation under each alternative.

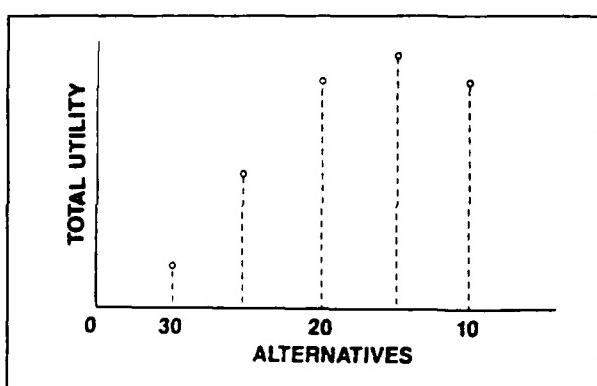


Figure 4. Relation of different plant control alternatives to total utility

Referring again to Figure 2 as the generalized case for total utility for a public good, as the level of consumption of the public good is further increased, total value rises to a maximum and then begins to decline. Past the maximum value, increases of the good actually produce decreasing levels of total utility; total value begins to decline with increased amounts of the good consumed.

This phenomenon is observed in natural resource management as the carrying capacity of a resource is exceeded. If the good is recreation access to open water for boating, additional boats on the lake can exceed the lake's carrying capacity, and congestion or unsafe conditions result, causing a decrease

in overall utility. Alternately, the productivity of fishery habitat could be easily exceeded.

In terms of aquatic plant control, the level of plants may decline so much that habitat values decrease and the fishery stock is adversely affected.

Demand: Specifying quantity-utility relationships

A demand curve relating public values to the amount of a public good provides useful information that supports decision-making. In developing such a demand curve for aquatic plant control, one notes that the two extremes of the curve can be readily defined from observation. If lake users cannot get their boats in the water or have a difficult time navigating through the water (under conditions of no control), then demand is high and the initial control efforts are highly valued. These conditions are readily observable from public behavior. Where control efforts have increased to the point that fishing habitat is reduced and perceptions of "good fishing" success are diminished, or when water quality benefits of the plants are reduced, the public perceives that there is too much plant control.

While these perceptions of too much or too little control may in part be "perceptions" alone, they will translate into preferences for particular control levels accompanied by different willingness-to-pay values for different control alternatives.

With the ability to estimate the demand and value for the two extremes of control, attention is turned to the broad middle range of control. The practical question asked is, Where on the demand curve should the control stop, i.e., what is the optimal point for control? The midrange value-quantity relations are the most important to know from a management standpoint.

For a demand curve with an inflection point (a curve that rises to a point and flattens out), control past the inflection point would still increase total utility or satisfaction, but the in-

creases would be small for each additional unit of control. The question here would be, Is this small increase in utility worth the cost of providing the additional unit of control?, given that most of the satisfaction possible has already been achieved in the steeper part of the demand curve. The question is best answered by examining the marginal utility of aquatic plant control.

Marginal utility concepts

To identify the point at which increases in plant control should be stopped requires examination of the marginal utility relationships (Figure 5). Marginal utility is the change in total value resulting from consumption of an additional unit of the public good or service. Marginal utility exhibits the phenomenon of diminishing marginal returns (Mansfield 1991, Thompson 1991). At some level or quantity of the good or service, the marginal utility will be zero. Further incremental increases in consumption actually result in decreases in the total utility.

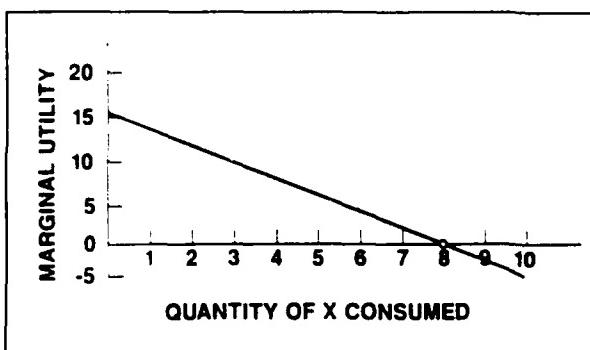


Figure 5. Relation of consumption of public goods to marginal utility

Analysis of the marginal utility function helps identify where additional plant control ceases to increase the overall value to the consumer. The amount of aquatic plant control that should be provided is that amount such that the marginal utility of the last unit of control is equal to zero. Less control than that amount results in benefits that could still be obtained by increasing the amount of control. Control levels past that point result in losses to total utility because the marginal utility of the additional control is negative.

Because provision of public goods and services is normally the responsibility of public and quasi-public entities, these questions about the marginal utility and midrange values of public good demand curves are more than theoretical economic constructs. Stating that a given level of plant control should be increased or decreased is best supported by being able to quantitatively show that a higher level will increase public benefits, or conversely, that total utility will not be diminished past a certain point, and that additional control will not increase total utility.

Valuation of Environmental Goods and Services

Determining the value of aquatic plant control efforts requires examining the value of the public goods and services described above and determining the value of affected private goods and services. Changes in aquatic plant infestation affect land values, hydropower production, agricultural production, commercial navigation, and industrial development, because of their dependence on a reliable source of water. These economic changes provide goods and services that are traded in a market, accruing benefits to private individuals and interests. Market data are available to support analysis of aquatic plant effects on the value of these market goods and services. The recreation and other public goods affected by aquatic plants and control efforts are not traded in markets, and thus the value of these services is determined through nonmarket techniques.

Market methods for environmental goods and services

Management of natural resources results in the production of products that have direct market prices, e.g., agricultural products, and in changes in the value of goods and services that are traded in a market, e.g. land values. Changes in environmental quality, resources, and natural resource characteristics lead to changes in production and variable and marginal costs, resulting in changes in the price and quantity of goods produced.

Market methods for valuation of environmental goods and services include direct valuation and indirect or surrogate market techniques. Agriculture or aquaculture production is the most obvious direct valuation process for a natural resource. The price of agriculture or aquaculture production changes with costs of production resulting from the effects of aquatic plants or aquatic plant control. Additionally, the value of various agency programs related to agriculture, forestry, or commercial fisheries can be directly measured as the change in total value of the market, resulting from increased productivity (Hufschmidt et al. 1983).

Another direct market method is the use of opportunity costs. The concept of opportunity costs is used most appropriately to value preservation of natural resources. The premise is that the value of the environmental characteristic or resource is equal to the sum of the foregone market values of the goods and services that are not produced because of preservation. This approach measures what has to be given up for the sake of preservation or nonuse. It does not measure the benefits of preservation for the unpriced uses (Hufschmidt et al. 1983). Perhaps the best utility of this concept may be in using opportunity costs to develop trade-off analyses comparing economic benefits to the benefits of preservation, e.g., habitat or historic values.

Surrogate market techniques include those methods that use market data, e.g. land values or travel costs, indirectly to determine values for environmental goods and services. Surrogate techniques include a number of land value approaches. Land value methods take the value of a fixed asset to be equal to the "discounted present value of the future net-benefits associated with the use of the asset" (Hufschmidt et al. 1983). The characteristics or attributes associated with the property are used to analyze the importance of the characteristics to the property's value.

The hedonic price approach and other land value applications assume that information on the demand for public goods is embedded

in the price and consumption levels of private goods (Freeman 1979). The value of property is used to determine the willingness-to-pay for different environmental characteristics. At Lake Guntersville, a hedonic analysis is being used to determine the impact of aquatic plant infestations on residential land values. Historic plant infestations are being compared to residential land values, as evidenced by home sales.

The Lake Guntersville work has illustrated some common problems associated with land value approaches. The data used for such an analysis are dependent on availability and quality of transaction data. The transaction data contain the records of home sales, length of time on the market, and other data that bear on the value of the home. Tax collection and real estate appraisal records provide information on the size of the residential lots, water frontage, and other home characteristics that affect the value of the property. All of these data have inherent biases and uncertainties. During the 15-year period of aquatic plant problems at Guntersville, long periods of relatively few residential transactions have occurred, due to other economic factors. This situation, coupled with property tax records that are not current, has provided a somewhat uneven data set. These considerations are common in such land value approaches.

The travel cost method is an additional surrogate method used for estimating recreation benefits. The method uses the individual's value of time and travel costs as a surrogate measure of willingness-to-pay for recreation. Although recreation has been the only application of travel cost analysis, other applications should be possible (Hufschmidt et al. 1983).

Nonmarket methods for environmental goods and services

For goods and services that cannot be valued directly or indirectly through market data, nonmarket valuation methods have been developed. Nonmarket methods entail establishing demand curves through bidding or voting methods to determine monetary val-

ues or to establish priorities among policies, outcomes, or alternative projects (Freeman 1979, Hufschmidt et al. 1983).

Bidding approaches are based on an individual's evaluation in paying for different bundles of goods and services. Willingness-to-pay values are elicited for improvements to an individual's situation, and willingness to accept compensation values for outcomes that reduce overall welfare. In contrast to the market methods, consumer preferences are determined for hypothetical situations rather than demand being based on observed consumer behavior.

At Lake Guntersville, valuation of different aquatic plant control alternatives is being undertaken through presentation of different control alternatives. Descriptions of recreation conditions to be experienced for each of three control alternatives will be presented. Respondents will provide two pieces of valuation information: their amount of recreation use and their willingness-to-pay for recreation under each of the control alternatives.

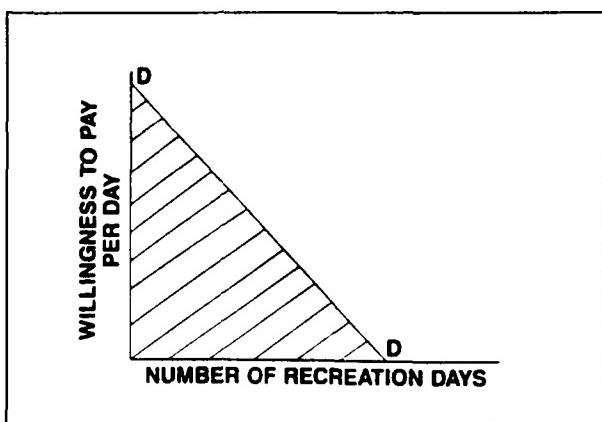
Establishing the relation between changes in environmental characteristics and valuation is more difficult for nonmarket analysis because of the lack of a functioning market to determine price. Freeman (1979) summarizes this process as establishing the change in three relationships. Application of this three-step process to the recreation analysis at Lake Guntersville gives the following:

- (1) Change in an environmental characteristic or amenity, e.g. level and distribution of aquatic plant control, leading to a change in measure of environmental quality. At Lake Guntersville, this is a change in level and distribution of aquatic plants (environmental characteristic), which leads to a
- (2) Change in environmental quality leading to a change in the flow of environmental goods and services. At Lake Guntersville, this is a change in

recreation quality and desired quantity (environmental goods and services), leading to a

- (3) Change in environmental services leading to changes in economic welfare. At Lake Guntersville, this is a change in willingness-to-pay for recreation use at Lake Guntersville (change in economic welfare or benefits).

Establishment of these relationships requires understanding of individual behavior and preferences under different conditions. The demand curves of Figure 2 represent aggregated preferences of a user population. To begin the process, say for recreation, a demand curve is determined for an individual showing the relation of willingness-to-pay to the number of recreation days consumed (Figure 6).



*Figure 6. Demand curve for recreation:
Relation of willingness-to-pay and
consumption of recreation days*

Individual willingness-to-pay values may be aggregated (Figure 7) to approximate an aggregate demand curve. In Figure 7, lines A, B, and C are the demand curves for individuals, and line D represents the aggregate or combined curve. The important point to note is that, in some cases, individuals hold greatly different willingness-to-pay values for the same level of an environmental amenity. Rather than individuals, A, B, and C could represent demand curves for different user groups and D, the total population demand curve. Where different groups are being considered,

the demand curves may show different preferences, i.e., downward-sloping versus upward-sloping, showing different preferences for overall levels of control.

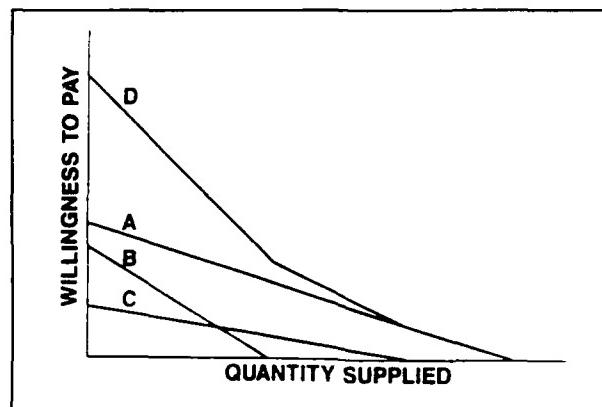


Figure 7. Aggregation of individual demand curves

The aggregation of user groups and individual demand curves can mask the differences in individual and group preferences. Generalized demand curves such as Figure 2 should be viewed in light of the understanding that an aggregated demand curve represents the combined demand of the individuals and groups represented.

Willingness-to-Pay Measurements

For goods and services traded in a market, the price is determined by the monetary amount that individuals will pay for a specified quantity of goods that suppliers are willing to supply at that price, shown graphically in Figure 8. The price represents the amount an individual is willing to pay for a particular quantity of the good. That is, the utility, degree of satisfaction, or level of economic welfare for individuals can be measured in terms of the prices they are prepared to pay for the consumption of goods and services. For goods that are consumed without paying for them, prices that individuals would be willing to pay can be imputed through nonmarket methods. For goods and services not traded in markets, what the individual has to pay to enjoy an environmental good, e.g. sportfishing, may be limited to his expenditures for a fishing license, tackle and equipment, and the value of his time.

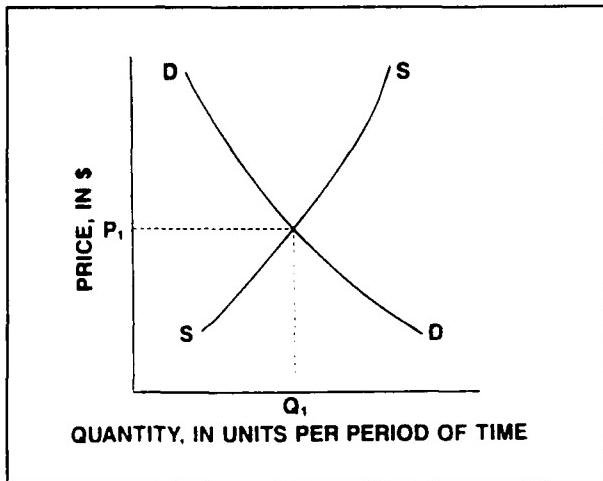


Figure 8. Price as a function of supply (S-S) and demand (D-D)

The provision of the sportfishing opportunity is supplied by a public agency, which bears most of the costs of providing the public good. This leads to a situation in which an individual's willingness-to-pay for a public good may far exceed what it costs the individual to consume the good. This relationship is shown in Figure 9, showing relation of expenditures to willingness-to-pay, and is normally referred to as a Marshallian demand curve.

This demand curve (Figure 9) shows willingness-to-pay related to quantity consumed, and the relation of expenditures to willingness-to-pay. The amount the individual is willing to pay but does not have to pay, i.e., the area under the demand line but above the expenditure line, is known as the consumer surplus. The consumer surplus is the difference between the amount the user is willing

to pay for a good and his actual expenditures for the good. Consumer surplus is considered the amount of public benefit produced by provision of the public good to the individual. Group and population measures of economic benefit are produced by aggregating the consumer surpluses of individuals in the user groups and population.

Agency actions that affect willingness-to-pay include such things as provision and quality of facilities, any required use or entrance fees, and the quality or condition of the natural resources and environmental amenities that are encountered in the consumption of the public good. Considering the willingness-to-pay for recreation affected by aquatic plants, willingness-to-pay may be reduced through excessive plant densities that clog boat motors and tangle water skis. Alternately, willingness-to-pay values may be increased by perceptions of improved angler success.

Development of willingness-to-pay models is accomplished through regression analysis that specifies the relation between willingness-to-pay and a number of explanatory variables that consider characteristics of the user, e.g. age, income, preferences for recreation, and variables that incorporate the aquatic plant conditions and other natural resource conditions (Freeman 1979). Analytical methods such as contingent valuation utilize this type of regression analysis to produce models that are used to value changes in recreation or other public goods.

Analysis Procedures

The economic benefits and costs that result from the operation of a public waterway are used to make decisions on the feasibility of construction of those facilities. While there may be generalized references to benefit-cost ratio or benefit-cost analysis or "having to take a look at the benefits versus the costs," such references may be without a specific conceptual framework. More importantly, the actual decision process that occurs may be misrepresented by the rather inexact use of economic terms.

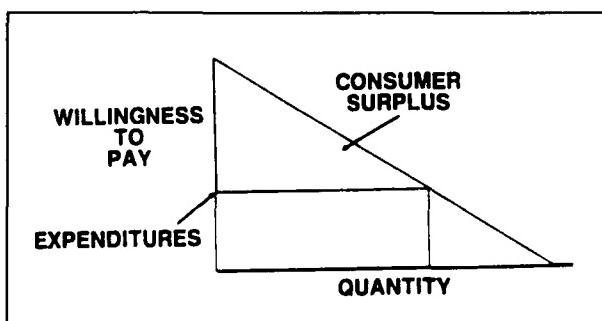


Figure 9. Consumer surplus

Cost-effectiveness analysis

Decisions about aquatic plant control consider the costs and benefits of control, but these decisions are by definition cost-effectiveness analyses rather than benefit-cost analyses. In cost-effectiveness, specific or target levels of goods or services are decided on as being desirable or the appropriate policy for adoption, and the cost-effectiveness analysis involves evaluating how to most effectively use available funding to achieve the goal levels (Hufschmidt et al. 1983).

As pointed out in the discussion on demand, the decisions on aquatic plant control involve specific levels of plant infestation or percent coverage to be achieved by proposed control alternatives. Given the available funding, decisions are made on how to allocate the funding to achieve the desired control alternative.

Estimating accurately the benefits and costs of public goods is more difficult than evaluating the most efficient way to achieve an agreed-on level of control (Hufschmidt et al. 1983). Obtaining a consensus of public and agency values for a particular level of aquatic plant control and then examining the available funding and other resources for attaining that level of control produces a cost-effectiveness analysis that can be used for decision-making.

A major difference with the benefit-cost analysis is the time horizon. Rather than considering benefits for the life of a project, annual decisions are made about control efforts. The nature of aquatic plant growth and redistribution is such that significant increases or decreases in plant biomass or acreages may occur in a single annual growing season. The effects of natural occurrences such as floods or droughts or human-controlled influences, such as reservoir drawdowns, can greatly change aquatic plant populations from year to year. For these reasons, an annual evaluation and planning of control efforts is required.

The cost-effectiveness criterion is used by decision-makers in formulating the plans for

control efforts. This decision process has been summarized as seeking to ascertain the level and distribution of control to be attained by available control technologies and other resources. This summary of the decision process will be examined in two parts (criteria) to more closely examine how economic information is used in the process.

Criterion: "Level and distribution of control." In the explanation of cost-effectiveness analysis, the discussion of determining the cost-effectiveness of available control plans to achieve a consensus control level may imply that an appropriate level of control is readily observed, obtained, or that consensus readily exists. This is not necessarily the case. Elicitation of such preference information on aquatic plant control preferences is usually limited to the public meetings in conjunction with a Master Plan for a plant control program, and may be limited to general input on plant control without valuation input.

In addition, plant control objectives are sometimes stated as reduction of the plants to some percentage of the waterway or to some historic level or biomass level. Objectives stated in such terms are difficult to use for eliciting public values because individuals have difficulty relating their use and values to biomass or percent coverage statements.

Recalling the willingness-to-pay curve for recreation (Figure 6), different groups and individuals hold different preferences for the same level of control, and these preferences for use may in fact conflict. To identify aquatic plant perceptions and preferences for different plant control levels, Lake Guntersville recreation users are being asked a series of questions. Information about the user's recreation use of the lake will allow aggregation of individual responses to analyze group preferences and values and to identify differences in plant control preferences and values between groups. The basic questions used to elicit this information are as follows:

Would you describe the aquatic plant coverage as:

- | | |
|-------------------|---------------|
| A. Not noticeable | D. Heavy |
| B. Slight | E. Severe |
| C. Moderate | F. Don't know |

What amount of aquatic plant coverage would you like to see?

- | |
|---|
| A. As much as possible |
| B. Somewhat more than presently exists |
| C. The same as presently exists |
| D. Less than presently exists but at least some |
| E. No coverage, eliminate the plants |
| F. Don't know |

In addition to the perception and preference questions, respondents provide information on issues and site characteristics, collectively called satisfactions and preferences, which are important for their recreation use. Such questions are a means to identify those externalities important for use by the recreationist. An abbreviated listing is given below.

Satisfactions and Preferences

- Open water where you can recreate
- Water quality
- Natural beauty
- Opportunity to catch large number of fish
- Public access to lake
- Habitat

As pointed out, plant control objectives stated as percent coverage or total biomass do not address an important aspect of the infestation that affects public preferences, that is, the distribution of the plants. To the individual user, the location of the plants may be more important to his willingness-to-pay than the total quantity or acreage coverage of the plants. Knowledge that a person's boat dock will not be blocked by plants will likely be more important to estimates of expected use and value than his knowing that a particu-

lar level of plant coverage equates to 27 percent of the reservoir.

Accommodating preferences on plant distributions has a number of technical aspects that must be considered. Response E to the second question above, "No coverage, eliminate the plants," while being a reasonable response, is impossible to accomplish from a biological standpoint, besides being undesirable for overall ecological considerations. Growth of most exotic aquatic plants is limited by the need for sufficient sunlight, normally limited by clarity of the water, but mostly by water depth. Only parts of a water body less than about 15 ft¹ in depth will support plant growth.

Water quality considerations restrict chemical treatment near water intakes. However, aquatic plants near major sources of agricultural runoff can absorb substantial amounts of excess nutrients from agricultural production. Considerations of lake recreation management support the need for open non-vegetated areas for water skiing, boat launching, and access to open-water areas.

Prior to formulation of alternatives for use in a valuation survey for Lake Guntersville, an effort was undertaken to obtain expert input on the distribution question. The intent was to ensure that the alternatives presented to the public are feasible and desirable and accommodate the diverse ecological and public needs of different user groups. A Technical Advisory Panel (TAP) was formed of five groups with expertise in the areas of aquatic plant management; recreation; water quality; fisheries; and wildlife, waterfowl, and wetlands.

The five expert groups worked separately to develop plant distributions that optimized benefits to their group's interests. After development of the five separate plant distribution plans, the groups prepared a TAP Plan representing a consensus plant distribution that accommodates all reservoir user groups.

¹ A table of factors for converting non-SI units of measurement to SI units is presented on page xxi.

Additionally, the plant distribution and acreage coverage of the TAP Plan approximated the goal of the Joint Agency Project of 7,000 acres of aquatic plants (10 percent of the reservoir).

Criterion: "Attained by available control technologies and other resources." The second part of the cost-effectiveness criterion subsumes all the constraints under which aquatic plant control occurs. A limited number of control technologies are available: physical control (reservoir drawdowns); biological control (establishment of natural predator populations); chemical treatment (herbicides); and mechanical control (plant harvester). The resources that affect decisions about aquatic plant control are subsumed under two headings, institutional factors and natural resource factors, as discussed below.

Cost-effectiveness decisions are made considering the institutional context of a bureaucratic agency. The most important institutional factor is available funding for plant control. Water quality and other regulatory institutional requirements will alter the level of aquatic plant control that is required. Considering the overall demand curve shown in Figure 2, if a level of control is decided, a higher level of control may be imposed (for instance, a Congressionally mandated goal as with Lake Guntersville). Such a newly established quantity of public goods produced will have the effect of increasing both total benefits and total costs. Depending on the starting point on the curve, the marginal benefit of the change may be less than the marginal costs of the required increase in control.

Changes in natural resources can alter the aquatic plant levels and distributions through changes in growing conditions. This will require alterations to the short- and long-range control plans. Referring to the demand curve, a drought can shift the total utility curve to the left, reducing benefits for the same amount of control effort. Alternately, flood years will increase the resulting control benefits realized.

Each control technology has different costs, length of effect, and public acceptability. Mechanical harvesters are expeditious in cutting boat lanes to ensure access to deep water, but may not be cost effective for small projects, because of the investment required. Biological controls have potential for the longest control period, but it may take several years for a population of the control organism to be large enough to provide the needed level of control. Difficulties in establishing the natural predators sometimes require several efforts. Chemical control is expedient, readily controlled, and cost effective, but has varying degrees of acceptability with the public.

Efforts to assess public perceptions on the different control techniques have been limited to public meeting input. The surveys at Lake Guntersville are eliciting perception information on the knowledge of and acceptability of different control techniques, and the effectiveness of the disseminated educational information. For a specific control decision, the costs of potential control technologies must be balanced with the benefits to be attained, the time extent of control, and public acceptability of the plant control project.

Summary

This paper has examined the economic valuation of a public good—control of nonnative aquatic plant species—through examination of methods to determine the economic value of the goods and services impacted by aquatic plant control. The approach used in the paper was to describe how the valuation could occur. Making decisions on plant control requires integration of economic benefit information with other policy and technical information. From the economic standpoint alone, the magnitude of this task can be shown by considering the tasks involved in optimizing the multiple effects and economic values, as shown in Table 1 (from Henderson 1991). The work accomplished in the Lake Guntersville study and other work at WES will increase the ability to value the benefits resulting from aquatic plant control efforts.

Table 1
Aquatic Plant Impacts on Economic Benefits

Project Purpose/Benefit	Aquatic Plant Impacts	Change in Benefits or Costs
Municipal and industrial water supply	Clogging of intakes; control efforts to avoid intakes.	Increased costs to supply; increased costs of control.
Flood control	Reduction of total storage capacity.	Potential loss of flood benefits of lost storage.
Hydropower	Clogging of intakes.	Increased generation and maintenance costs.
Inland navigation	Increase in difficulty of access to loading/unloading facilities.	Increased transportation costs.
Recreation	Affects components of recreation use.	Changes in recreation use such as fishery habitat, accessibility and navigation, and willingness-to-pay for recreation.
Regional income	Change in income due to impacts of plants on water-dependent industries.	Changes in income, e.g. recreation-related, manufacturing.
Regional employment	Changes due to impact of plants on water-dependent industries.	Changes in employment, e.g. recreation-related, manufacturing.
Urban and community impact	Changes in residential land use patterns and residents' use of the lake.	Changes in residential land values due to aquatic plants.
Life, health, and safety	Changes in vector production; possible hazards to safety of plants.	Losses due to vector-borne disease.

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Movements and Habitat Use of Triploid Grass Carp in Lake Marion, South Carolina, 1990-1991

by

Stephen D. Kartalia,¹ Jeffrey W. Foltz,¹ and K. Jack Killgore²

Introduction

Lake Marion, South Carolina, is a 44,000-ha lake formed by impoundment of the Santee River. It consists of open water as well as dense cypress swamps. Aquatic vegetation has become a serious problem in upper Lake Marion north of the Interstate 95 (I-95) bridge (Inabinette 1985). Upper Lake Marion's shallowness is conducive to aquatic plant growth. Many areas of the lake have limited access due to dense aquatic vegetation. This has hampered use of the lake by recreational hunters and anglers. Herbicides have been used extensively to control the vegetation problem, but a more feasible and long-term solution is needed. Mechanical removal and herbicide treatment can be costly and time consuming, and work on a limited basis for a limited time.

One possible solution is the release of triploid grass carp (*Ctenopharyngodon idella*). Grass carp have been effective in eliminating *Hydrilla verticillata* in several large lakes in Florida (Beach et al. 1976; Miley, Leslie, and Van Dyke 1979). Advantages of biological control include longevity of the method, constant fish-feeding activity against growing vegetation, low long-term cost, and high effectiveness on selected plants (Sutton and Vandiver 1986).

Three hundred thousand triploid grass carp have been released between 1989 and 1991 at various locations in upper Lake Marion to control nuisance aquatic vegetation (South Carolina Aquatic Plant Management Council and South Carolina Water Resources Com-

mission 1990). Water temperature is known to cause migrational movement in grass carp once water reaches 15 to 17 °C (Aliev 1976). A rise in water level or increased flow rates have also caused grass carp to exhibit migrational movement (Stanley, Miley, and Sutton 1978). If triploid grass carp were to migrate up the rivers and away from aquatic plant-infested areas, they would be ineffective for weed control.

Objectives of this study were to (1) determine the magnitude and direction of grass carp movements, (2) determine if grass carp remain in the targeted vegetation areas, and (3) examine characteristics of habitats used by triploid grass carp.

Study Site

Lake Marion has an average depth of only 5 m and a maximum depth of 12 m. Immediately upstream of Lake Marion, the Santee River is formed by the confluence of the Wateree and Congaree Rivers. The Wateree River originates at the Wateree Dam about 100 km upstream from Lake Marion.

The Congaree River originates at the Saluda Dam on Lake Murray and flows 85 km before joining the Wateree. The Wateree and Congaree Rivers average 183 and 266 m³/sec discharge, respectively. When Lake Marion was constructed in 1941, it impounded 6,500 ha in its headwater section, known as the Santee Swamp. The swamp is anaerobic most of the year and hence influences water quality in upper Lake Marion (Bates and Marcus 1989). Vegetated areas of the lake targeted for control

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² US Army Engineer Waterways Experiment Station, Vicksburg, MS.

have an estimated 4,800 ha of submerged vegetation, mostly upstream from the I-95 bridge.

Habitats in upper Lake Marion can be categorized as five types: (1) Santee River channel (2 to 8 m deep), (2) open water with creek channels running through it (1 to 2 m deep), (3) open water with scattered cypress trees (2 to 3 m deep), (4) open-water shallow flats (1 to 2 m deep), and (5) thick cypress swamp (2 to 3 m deep). All types except the Santee River channel support dense stands of nuisance aquatic vegetation. No assessment of the proportion of upper Lake Marion that each habitat type comprises has been made. Likewise, no assessment of the magnitude of the swamp's influence on downstream water quality has been made, although fish kills due to anaerobic conditions are a common summer phenomenon in upper Lake Marion's swamps.

Methods

Thirty-seven triploid grass carp were surgically implanted with radio transmitters. The life span of the transmitters was 9 months, and transmitter frequencies ranged from 48.036 to 49.527 KHz. Each fish was identified by a distinct frequency. Fish were anesthetized using a bath containing 100 mg/L MS-222 and 25 mg/L Furacin. Each fish was weighed to the nearest 0.01 kg and measured to the nearest millimeter. Anesthetized fish were placed in a V-shaped operating trough so that the fish excluding its abdomen was submerged in water containing MS-222 and Furacin. A small aquarium aerator was used to maintain adequate oxygen levels in the operating trough. A radio transmitter was then surgically implanted using the procedure described by Schramm and Black (1984). Surgical gloves were worn, and instruments and transmitters were disinfected prior to use.

Scales were removed from the incision area, and a 5-cm longitudinal incision was made in the ventral wall 6 cm anterior to the pelvic girdle. A transmitter was inserted into the body cavity, and the incision was closed with nonabsorbable silk sutures. Oxytetracycline (50 mg/kg body weight) was injected into the body cavity before the last suture was stitched. Fish were then immediately released into the lake approximately 1 km north of Santee State Park (Figure 1).

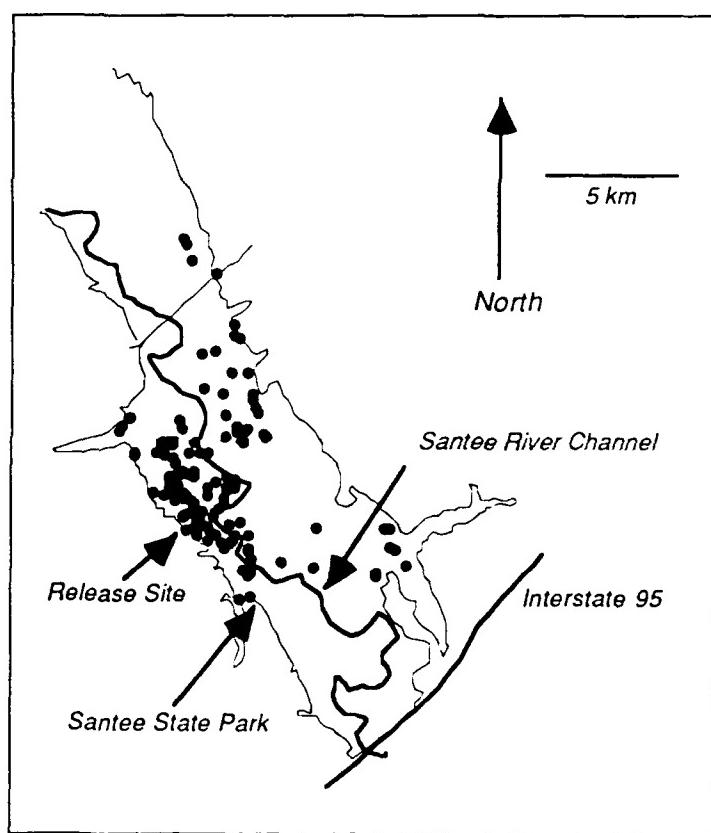


Figure 1. Locations of radio-tagged adult triploid grass carp in upper Lake Marion, 1990-1991. Each dot represents one or more individual fish locations

An Advanced Telemetry Systems (model 2000) radio receiver was used. Boat searches for implanted grass carp were conducted 3 days per week for 18 months. Signals were received while boating with a Telex Communications (model 64 B-S) four-element yagi antenna. Once a signal was picked up by the

receiver, the antenna was rotated to ascertain direction, and the boat was motored in that direction. Signal strength increased as the fish was approached. The coax cable was then disconnected from the antenna and dropped in the water beside the boat. Intensity of the signal indicated when the boat was within 25 m of the fish.

Once a fish was located, the date, water depth, and Loran latitude and longitude coordinates were recorded. Water temperature and dissolved oxygen (DO) were measured at the surface and on the bottom with a YSI dissolved oxygen meter (model 51B) (to the nearest 0.1 °C and 0.1 mg/L, respectively). Mean DO was calculated from the surface and bottom value. An aquatic vegetation sample was taken from the surface and bottom with a rake. Aquatic vegetation was identified (Pennwalt 1984) in the field. The species that comprised the largest proportion of a sample was categorized as primary vegetation, and the species that comprised the next largest proportion was categorized as secondary.

Vegetation density in the general vicinity of the fish location was identified as one of four categories: (1) vegetation covers ≥50 percent of the surface, (2) vegetation covers <50 percent of the surface, (3) vegetation present but submersed, or (4) vegetation sparse. Habitat type was categorized as one of five types (see **Study Site**): (1) river channel (Santee, Congaree, or Wateree); (2) open water with creek channels; (3) open-water shallow flats; (4) thick cypress swamp; and (5) open water with scattered cypress trees.

Fish locations were plotted on a digitized map that indicated the Santee River channel. Days elapsed and distance moved (to nearest 0.01 km) between readings were computed for each fish. Minimum net daily movement was then computed as net kilometers per elapsed days. Linear and nonlinear regressions were employed to describe seasonal changes in minimum net daily movement. Distance from the river channel to the fish was computed along a line perpendicular to the fish and the river channel. Distances

were recorded to the nearest 0.01 km. Distance of grass carp from the river channel was tested with a t-test (null hypothesis that distance = zero).

Results

Triploid grass carp used in this study averaged 704 mm total length (standard error, SE = 15) and 4.42 kg live weight (SE = 0.20) at the time of release. Fish were located on 180 occasions, and the average elapsed time between locations of an individual fish was 17 days. The longest distance moved by a fish was 10.6 km over 4 days, while the average was 0.10 km/day (SE = 0.01). Grass carp in this study did not demonstrate a preference for the river channel in Lake Marion (Figure 1). Mean distance of grass carp from the river channel was 1.01 km (SE = 0.07). This mean distance was significantly different from zero ($t = 14.21$, $p = 0.0001$).

Surface DO concentrations at fish locations remained above 8 mg/L most of the year, but bottom concentrations averaged less than 1 mg/L in May (Figure 2). Water temperatures at fish locations were similar to the pattern that occurs in the upper lake: winter temperatures of 10 °C and summer high temperatures of 29 °C (Figure 2). There was a 2 to 5 °C difference between surface and bottom temperatures at fish locations, which ranged in depth from 2 to 3 m (Figure 2).

No studies to date have quantified the different proportions of habitat and aquatic vegetation types in upper Lake Marion. During the study, 64 percent of fish locations were habitats composed of shallow flats with depths of 2 to 3 m (Figure 3). Twenty-five percent of the time, grass carp locations were in thick cypress swamps. Fish were congregated in either open-water shallow flats or thick cypress swamps during the months of January through April (Table 1). Numbers of fish in the cypress swamps decreased from June through September, and numbers of fish in shallow flats increased. No fish were located in the cypress swamp by June.

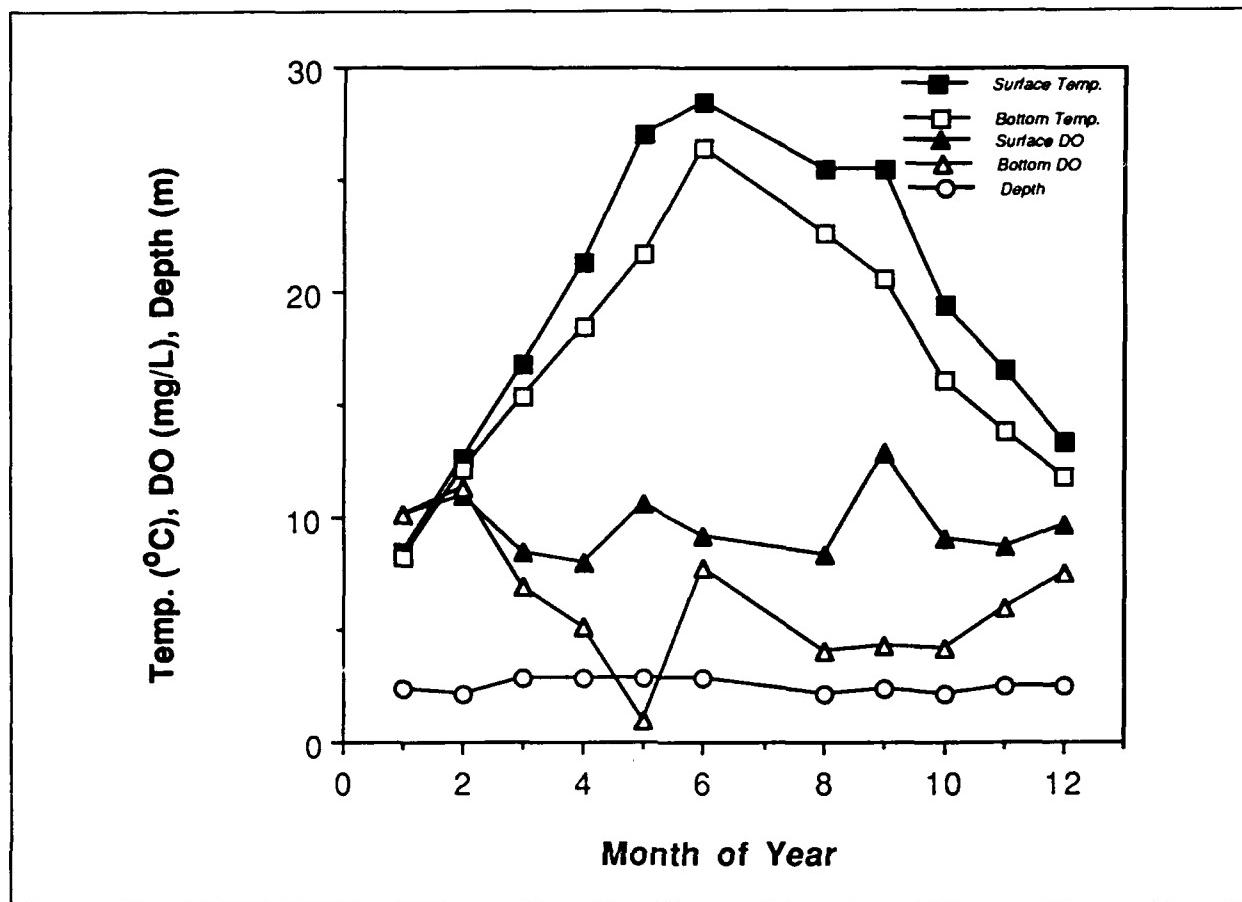


Figure 2. Average depth (m), water temperature ($^{\circ}\text{C}$), and dissolved oxygen (mg/L) at locations occupied by adult radio-tagged triploid grass carp in upper Lake Marion

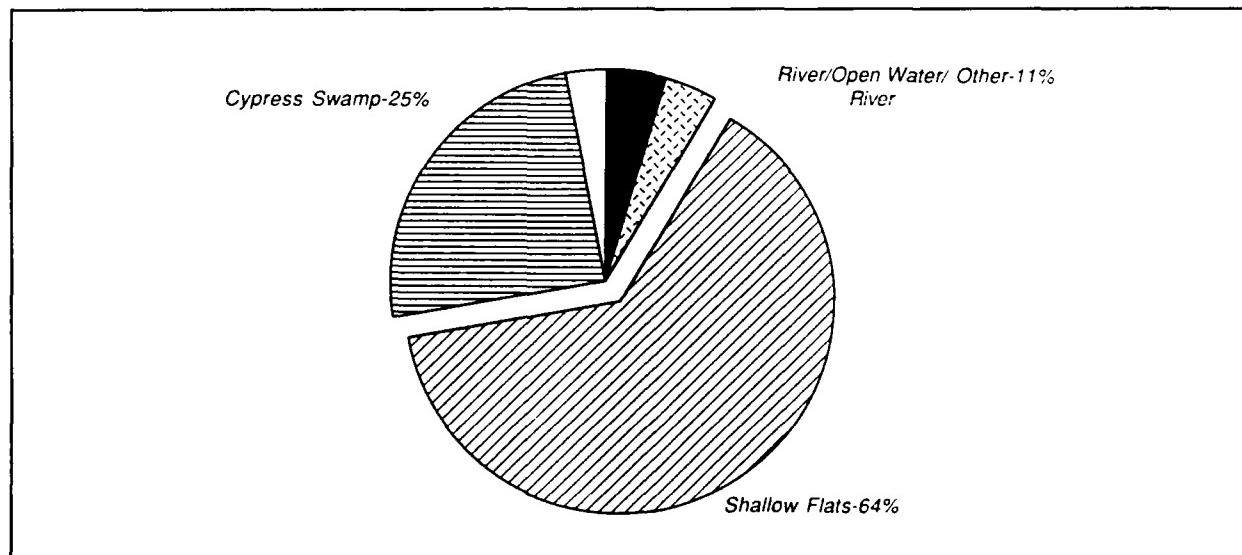


Figure 3. Habitat utilization by radio-tagged adult triploid grass carp in upper Lake Marion

Table 1
Percentage of Grass Carp Locations in Relation to Habitat Categories in Lake Marion

Month (1991)	Habitat Category (Percentage Fish Locations) ¹				
	RC	OWCC	OWSF	TCS	OWCS
January	0	0	57	43	0
February	0	4	48	48	0
March	0	0	71	26	3
April	0	0	84	16	0
May	0	13	75	12	0
June	0	25	75	0	0
July	100 ²	0	0	0	0
August	0	0	75	25	0
September	0	0	100	0	0
October	0	0	52	24	24
November	0	8	72	20	0
December	0	0	55	45	0

¹ Habitat categories are defined as follows: RC = river channel, OWCC = open water with creek channels, OWSF = open-water shallow flats, TCS = thick sypress swamps, and OWCS = open-water cypress stands.

² These fish were located at boundary between river channel and shallow flats near Santee State Park.

Fifty-three percent of recorded grass carp locations were in areas with aquatic vegetation at the water's surface (Figure 4). More specifically, 23 percent of the locations were areas with vegetation that covered ≥ 50 percent of the surface and 30 percent in areas with vegetation that covered <50 percent of the surface (Figure 4). Areas with submersed vegetation accounted for 41 percent of fish locations.

On a seasonal basis, grass carp locations demonstrated a trend similar to that of the progressive increase in aquatic plant biomass. The summer is characterized by dense aquatic vegetation stands that reach the water surface. Grass carp locations in areas rated as ≥ 50 percent coverage increased from 13 percent in May, to 75 percent in September, and declined to 43, 44, 55, and 0 percent for October,

November, December, and January, respectively (Table 2). Winter and spring frequency of observations for areas with <50 percent coverage and submersed vegetation were high, as expected, accounting for 100, 87, 91, and 100 percent of grass carp locations for January, February, March, and April.

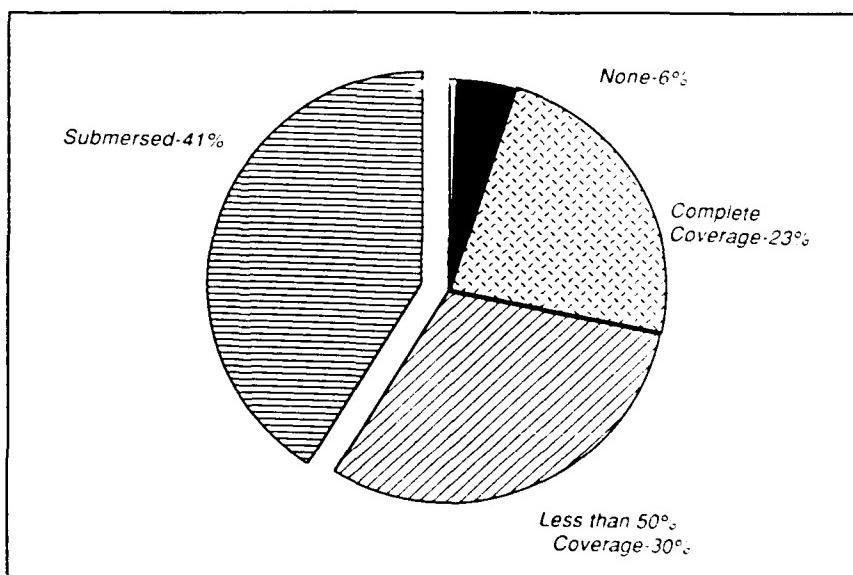


Figure 4. Aquatic vegetation densities of locations used by radio-tagged adult triploid grass carp in upper Lake Marion

Table 2
Monthly Changes in Vegetation Density at Grass Carp Locations

Month (1991)	Density Category (Percentage of Fish Locations)				
	None	Sparse	Submersed	<50% coverage	≥50-100% Coverage
January	0	0	71	29	0
February	0	0	30	57	13
March	0	3	74	17	6
April	0	0	79	21	0
May	0	0	38	50	12
June	25	0	75	0	0
July	100 ¹	0	0	0	0
August	0	0	0	38	62
September	0	0	0	25	75
October	5	0	38	14	43
November	0	0	4	52	44
December	0	0	18	27	55

¹ These fish were located at boundary between river channel and shallow flats near Santee State Park.

respectively. Over the course of the study, 70 percent of fish locations were in areas dominated by hydrilla (Figure 5). *Egeria densa* was the predominant vegetation type in only 6 percent of fish locations. Other vegetation types, which included duckweed, *Nitella*, and coontail, accounted for 19 percent of the locations, but no single species exceeded 2 percent.

Discussion

The magnitude of grass carp movements noted in this study was comparable but less than rates reported for adult fish by Chappellear et al. (1990) and Bain et al. (1990). Chappellear et al. (1990) reported average daily movements of 0.29 km/day with an average elapsed time between observations of an individual fish of 10 days. Movements reported by Chappellear et al. (1990) were for grass carp released at three widely separated points in upper Lake Marion. Also, fish probably moved to avoid the widespread low-DO events that occurred during 1989 and 1990 in the uppermost cypress swamps and adjacent flats. Bain et al. (1990) reported that adult fish movement averaged 33 km over a 4-month

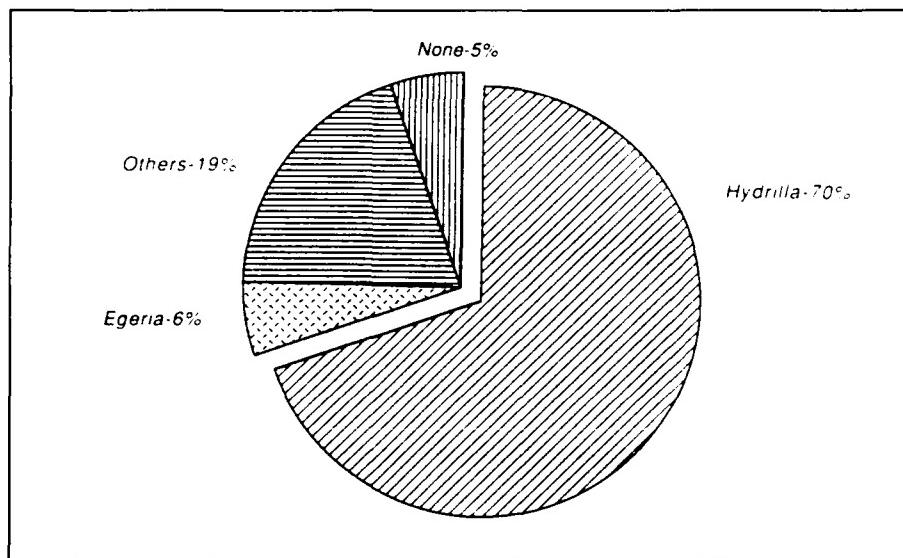


Figure 5. Predominant aquatic vegetation at locations used by radio-tagged adult triploid grass carp in upper Lake Marion

period (i.e., about 0.27 km/day) and that one fish traveled 6 km/day. It is difficult to make direct comparisons between studies without knowledge of the frequency of observation. For example, a fish could travel 1 km each day for 10 successive days, but if it finished at its origin and had not been observed for 10 days, net daily movement would compute as zero. In the present study, grass carp generally remained in the shallow flats located within 2 km of their release site.

Redistribution of grass carp from thick cypress swamps to open-water shallow flats was probably due to low DO in the upper lake during the summer. Thick cypress swamps are characterized by low summer DO concentrations (Bates and Marcus 1989). Utilization of slightly deeper and slightly less vegetated areas probably provides grass carp with a suitable combination of food density and DO concentrations.

Locations predominated by hydrilla constituted the majority of grass carp locations. Hydrilla is an excellent food for grass carp because of the soft nature of the plant and its high ash content (Tan 1970, Rottman 1977). Grass carp used in this study were large (704 mm total length), and according to Sutton and Vandiver (1986), hydrilla would be their preferred food. No data exist concerning the percentage of Lake Marion's total nuisance aquatic plants that the individual species comprise. Thus, no preferences for hydrilla can be inferred in the present study.

In summary, no long-distance migrations were observed, and fish showed no affinity for the Santee River channel. Triploid grass carp remained in the upper part of Lake Marion, and these fish did not leave areas targeted for aquatic vegetation control. Dissolved oxygen levels appeared to play an important part in the location and movement of fish. In addition, information on water quality and aquatic vegetation distribution for upper Lake Marion is needed in order to interpret movements relative to available habitat.

Acknowledgments

We wish to thank the staff at the South Carolina Wildlife and Marine Resources Department's Dennis Wildlife Center for their help and use of facilities and the South Carolina Water Resources Commission for their support.

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Management of Endemic Aquatic Macrophytes in Warmwater Ponds

by

Gary O. Dick¹ and R. Michael Smart¹

Introduction

The Lewisville Aquatic Ecosystem Research Facility, located in Lewisville, TX, consists in part of 55 earthen ponds and was originally operated as a state fish hatchery. All ponds are gravity-fed from Lewisville Lake and are inhabited by several species of submersed aquatic macrophytes, including *Najas guadalupensis*, *Potamogeton nodosus*, *P. pectinatus*, *Ceratophyllum demersum*, *Zannichellia palustris*, and *Chara vulgaris*. Additionally, numerous emergent macrophyte species are indigenous to the ponds.

To make the ponds more useful for controlled aquatic plant research, we require practical methods for reducing or eliminating the dominant species, *Najas* and *Chara*. Our initial approach has been to investigate the germination of *Najas* seed and *Chara* spore banks in pond sediment, the efficacy of water-level manipulation for reduction of seed and spore numbers, and the use of selected herbicides for preemergent control of both species.

Methods

Seed and spore bank germination

Sediment was collected from several ponds, and then pooled and mixed to ensure homogeneity. Aliquots of 0.5 L of sediment were added to 3.8-L pots (to a depth of 6 cm), and each pot was then flooded with 2.0 L of filtered (0.1 mm) lake water. Water temperatures were maintained between 20 and 30 °C

for 38 weeks under a 14:10 artificial light regime. Germinated plants were counted and removed on a weekly basis as soon as they could be identified as *Najas*, *Chara*, or "other." After germination rates tapered off (week 8), water from a set of pots was drawn off, and the sediment was dried for 1 week. Each pot of the set was then reflooded with 2.0 L of deionized water. The drying process was repeated at 25 weeks on these pots.

An additional set of pots was dried and reflooded for the first time at week 25, resulting in three treatments in the experiment: (1) continually flooded, (2) two dry periods (at 8 and 25 weeks), and (3) one dry period (at 25 weeks). The data presented here represent 38 weeks of observation. We plan to continue this study until the seed/spore banks are depleted.

Wet/dry cycles in ponds

Several ponds at the Lewisville Facility were manipulated to observe the effects of wet/dry cycles on *Najas* and *Chara* populations. Water levels were maintained for a minimum of 6 weeks after filling a pond, which allowed time for the initial surge of germination by *Najas* and most of the first surge by *Chara*. The ponds were then drained to destroy all germinated plants. After a drying period, the ponds were flooded again and the cycle repeated. Effectiveness was estimated visually by species composition and area of coverage by all species from cycle to cycle. Although these methods await scientific scrutiny, we present several observations related to this procedure.

¹ US Army Engineer Waterways Experiment Station, Lewisville Aquatic Ecosystem Research Facility, Lewisville, TX.

Preemergent control

Three herbicides were chosen based upon preliminary testing and/or known efficacies against the target species: monoamine salt of endothall, dichlobenil, and metam-sodium. Methods used in the germination study were duplicated. Sediment in each treatment was amended with maximum label-recommended rates of the herbicides for controlling standing macrophytes or seeds.

Results and Discussion

Seed and spore bank germination

Najas began sprouting approximately 1 week after flooding. Initial numbers averaged five germinations per replicate over the first 4 weeks of the study (Figure 1). *Chara* began sprouting approximately 2 weeks after flooding, with high numbers of plants removed during the first 8 weeks of the study (Figure 2). "Other" plant species, principally *Polygonum* spp., began sprouting 1 week after flooding, but generally died prior to harvest.

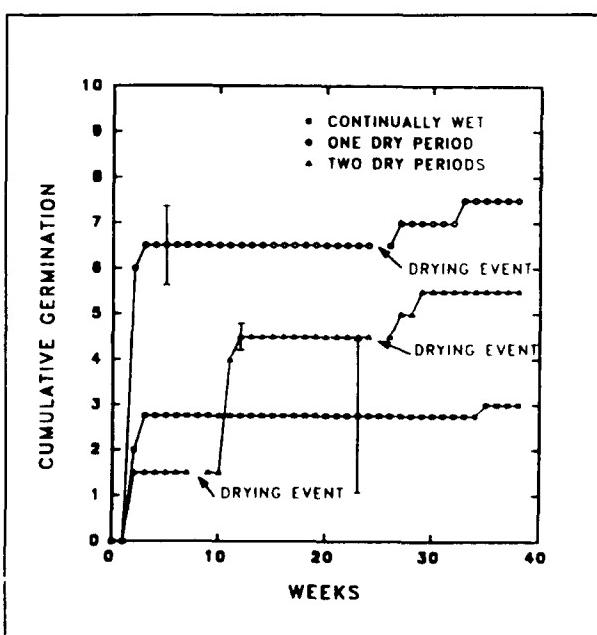


Figure 1. Mean cumulative germination of *Najas guadalupensis* in three treatments of wet/dry cycles, with a typical standard error bar for each treatment

Following the initial surge, germination rates tapered off in both species. *Najas* stopped sprouting after 4 weeks. *Chara* continued to germinate over the remainder of the study, but at much reduced rates after 8 weeks, with weekly averages of less than one plant per replicate in continually flooded pots.

The drying events at 8 and 25 weeks apparently triggered additional *Najas* seed germination, although cumulative numbers were not significantly different from the continually flooded treatment. Small sample size with relatively low germination numbers hampered our ability to detect differences between treatments.

The induced wet/dry cycles sparked increases in germination by *Chara*. The treatment dried at 8 weeks showed a significant increase over the continually flooded treatment, increasing the cumulative number of germinations by over 50 percent. Both treatments dried at 25 weeks showed less than 5-percent increases in total numbers of germinating spores. The large difference in the increases between the initial drying periods for the two wet/dry treatments may have been

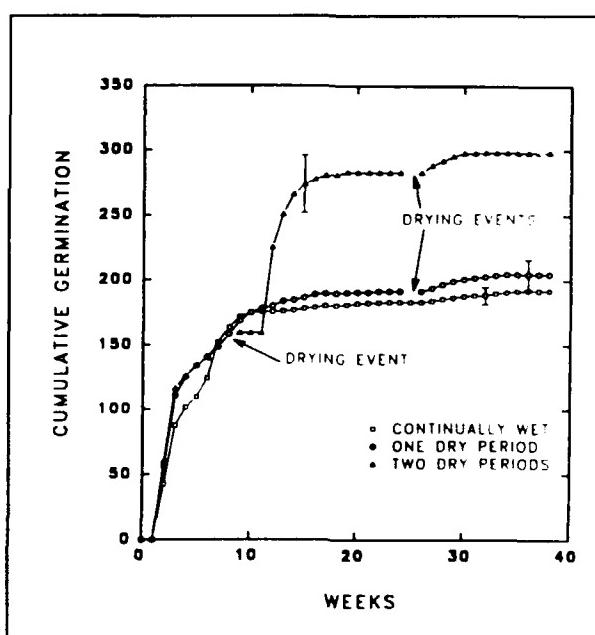


Figure 2. Mean cumulative germination of *Chara vulgaris* in three treatments of wet/dry cycles, with a typical standard error bar for each treatment

related to timing. Those spores that were dried early and reflooded may not have had an opportunity to enter into "long-term" dormancy, whereas those not subjected to an early dry period did, or were not able to survive.

Wet/dry cycles in ponds

In ponds that were drained and filled only once, *Chara* returned as the initial dominant species, but was gradually replaced by *Najas* when water was maintained in the pond for more than 10 weeks. This may have been the result of the high number of *Chara* spores germinating (estimated at >8,500/m² in some ponds) and the favorable conditions for *Chara* growth in newly flooded ponds (high levels of light and nutrients). Successional replacement by *Najas* over time may have been typical, due in part to shading and eventual crowding out of the *Chara*.

When ponds were subjected to repetitive short-term wet/dry cycles, *Najas* replaced *Chara* as the initial dominant species in second and successive fill periods, with *Chara* becoming established only in fringe areas where *Najas* did not grow well, usually in very shallow water. The first wet/dry cycle may have significantly reduced the spore bank of *Chara*, giving *Najas* an opportunity to become established earlier due to reduced competition.

Six weeks was the preferred length of time for the wet period of the cycle. Under longer periods, *Chara* may produce spores and negate the operation. Although *Najas* does not reach seed production potential as quickly, it is more resistant to dry periods and may require up to 2 weeks for complete elimination of plants by drawdown. A near-optimum wet/dry cycle thus requires 6 to 8 weeks.

Preemergent control

The numbers of *Najas* and *Chara* germinated from sediment treated with monoamine salt of endothall did not vary significantly from controls (Figures 3 and 4). Endothall

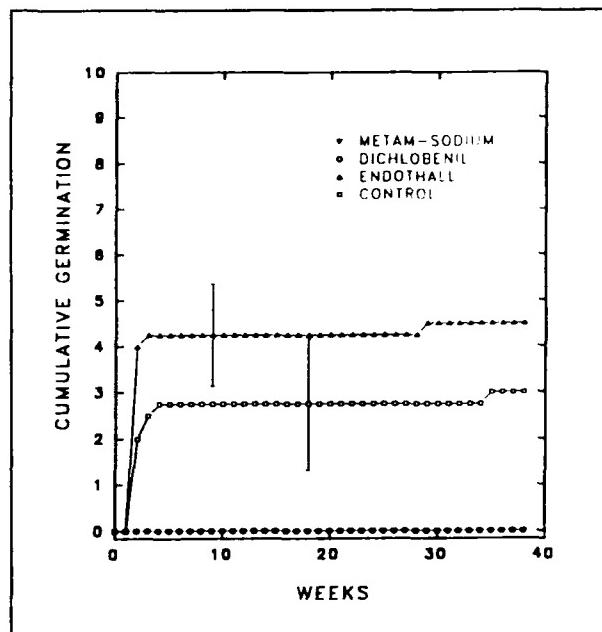


Figure 3. Mean cumulative germination of *Najas guadalupensis* in three treatments of herbicides, with a typical standard error bar for each treatment

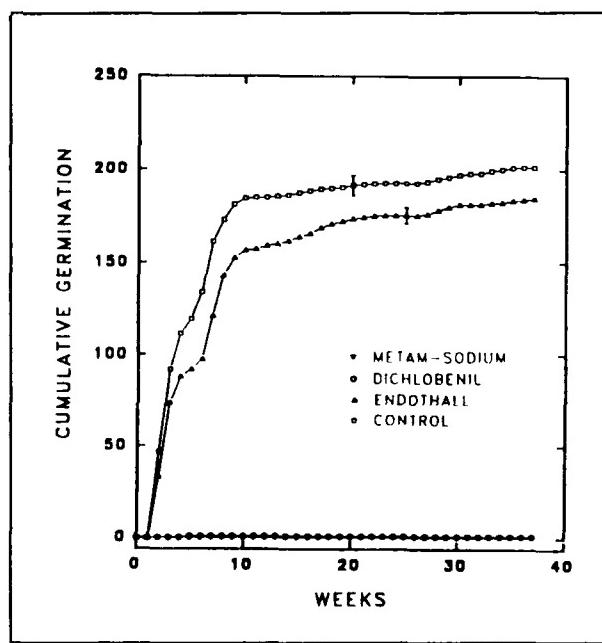


Figure 4. Mean cumulative germination of *Chara vulgaris* in three treatments of herbicides, with a typical standard error bar for each treatment

was deemed inadequate as a preemergent herbicide for use in ponds.

Dichlobenil and metam-sodium treatments showed significantly lower germination rates than controls, totaling only four individual germinated *Chara* spores and no germinated *Najas* seeds in dichlobenil treatments, and one germinated *Chara* spore and no germinated *Najas* seeds in metam-sodium treatments. The four *Chara* plants in dichlobenil treatments occurred in the third week of the study, and all died within 3 days.

Although dichlobenil was effective at controlling germination in both species, its use may be limited to ponds in which no aquatic plant studies are planned in the near future. The herbicide may persist in the sediment for up to 1 year, and nonselectively controls

standing macrophytes. We are continuing our evaluation of this compound for control of unwanted vegetation in experimental ponds.

Metam-sodium showed the most promise in preemergent control of *Najas* and *Chara*. When used as a soil fumigant, the herbicide kills both seeds and spores, effectively sterilizing the sediment for our purposes. After several days of drying, the metam-sodium vapors escape the sediment, and preliminary re-vegetation tests on treated sediment thus far have been very successful. Further study involving application techniques on a larger scale is currently being planned.

Effects of Aquatic Plants on Water Quality in Pond Ecosystems

by

David Honnold,¹ John D. Madsen,¹ and R. Michael Smart¹

Introduction

This study was undertaken to directly compare the effects of exotic plant species versus native plant communities on water quality. To date, the relative influence of exotic aquatic plant species on water quality has received little attention. A series of culture and research ponds located at the Lewisville Aquatic Ecosystem Research Facility (LAERF) provided a unique opportunity to examine the impacts of different aquatic plant species on water of similar origin. Ten ponds of different species composition were examined at various times during the study. In this effort, we used continuous hourly recordings of water quality parameters to provide more complete observations of daily variations than the single-point observations often used.

The goal of this study was to document both diurnal and longer term patterns in four water quality parameters (temperature, dissolved oxygen, pH, and conductivity) in ponds of different plant composition.

Methods

Vegetation types selected for this study included the exotic species of waterhyacinth (*Eichhornia crassipes*), hydrilla (*Hydrilla verticillata*), Eurasian watermilfoil (*Myriophyllum spicatum*), and a native species complex that included American pondweed (*Potamogeton nodosus*), southern naiad (*Najas guadelupensis*), coontail (*Cerato-*

phyllum demersum), and stonewort (*Chara* sp.). Three ponds of waterhyacinth (floating) were used with a total sampling period of 53 days. One pond of hydrilla (submersed) was used with a total sampling period of 47 days. Four ponds of Eurasian watermilfoil (submersed) were used with a total sampling period of 41 days. Two ponds containing various native species (submersed) were sampled with a total sampling period of 53 days.

The average size of the study ponds is 0.30 ha, with an average depth of approximately 1 m (Figure 1).

Four Hydrolab Datasonde I units were calibrated (as outlined in the manual) and deployed on a weekly basis, rotating between ponds of each species composition. Each sonde deployed was located in the deepest area of the pond, and was positioned such that the probes were located subsurface. One pond per representative species composition was monitored each week.

Parameters measured were temperature (°C), dissolved oxygen (mg/L), pH (units), and conductivity (µS/cm). All conductance values were corrected to 25 °C. The recording sample frequencies were set at 1-hr intervals.

Data analysis was undertaken using a Kruskal-Wallis nonparametric statistic. The statistics were computed for the minimum, maximum, average, and range for each 24-hr day of recorded information for each pond type.

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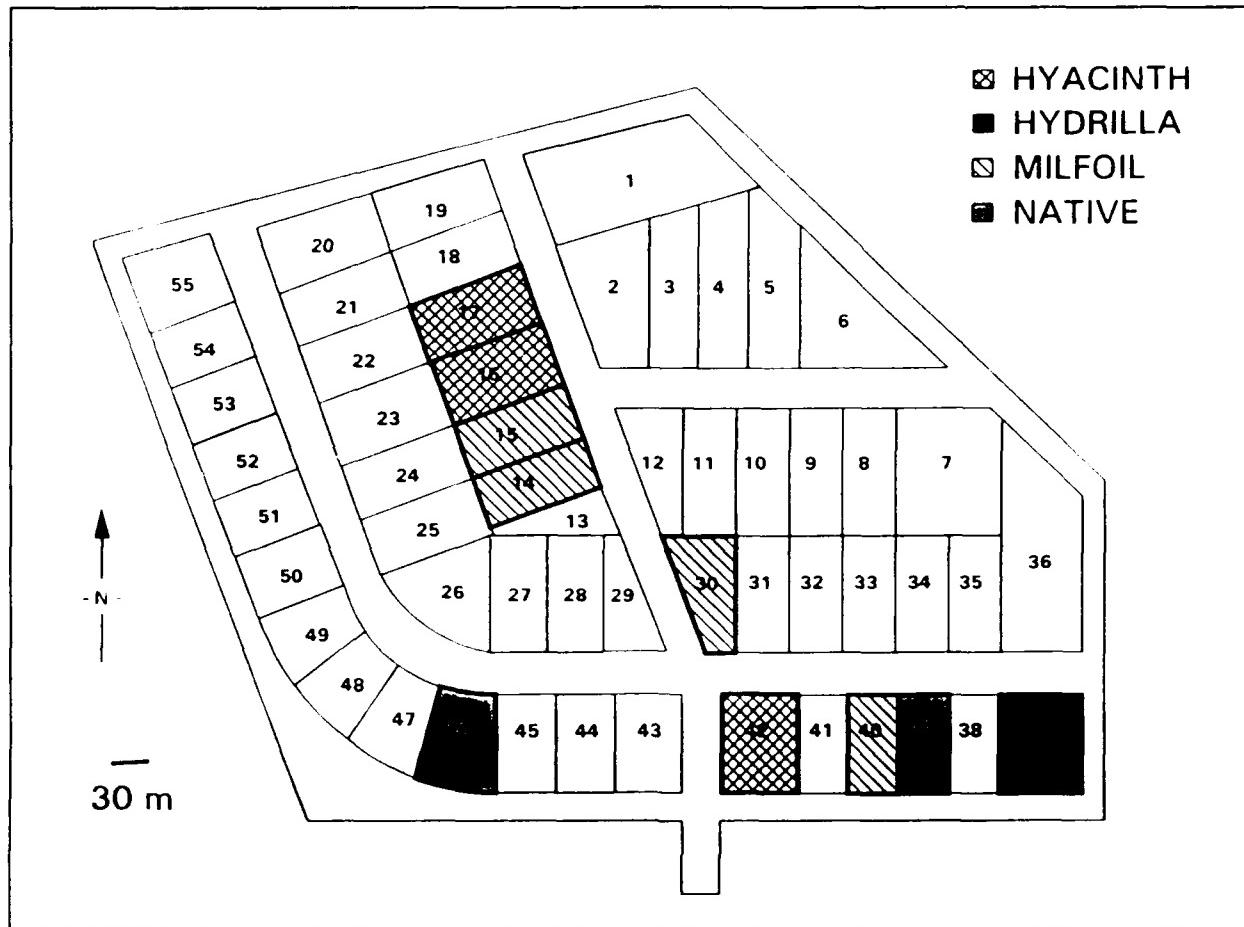


Figure 1. Map of Lewisville Aquatic Ecosystem Research Facility showing pond locations, size, and species composition

Results and Discussion

Surface water temperature of the ponds revealed very little difference in daily average values (Figure 2).

Dissolved oxygen in the hydrilla ponds was significantly lower than in the watermilfoil or native ponds (Figure 3). Dissolved oxygen was highest in the native species ponds. Average dissolved oxygen levels in the exotic species ponds were below 5 mg/L, which is undesirable for warmwater fisheries.¹ The daily fluctuations of dissolved oxygen were similar in amplitude, but the minimum daily values observed in ponds containing exotic species could cause stress to warmwater fish.

Average pH values were highest in the native ponds, lowest in the waterhyacinth ponds, and intermediate in the hydrilla and watermilfoil ponds (Figure 4). Daily fluctuations in pH were fairly large in the submersed species ponds, while pH of the waterhyacinth (floating) ponds exhibited little daily variation.

Conductivity values among the exotic species ponds were very similar, and exhibited little daily variation (Figure 5). The native ponds exhibited both lower conductivity values and greater diurnal fluctuations than did the exotic species ponds.

¹ Claude E. Boyd. 1979. Water quality in warmwater fish ponds. Auburn, AL: Auburn University, Agricultural Experiment Station.

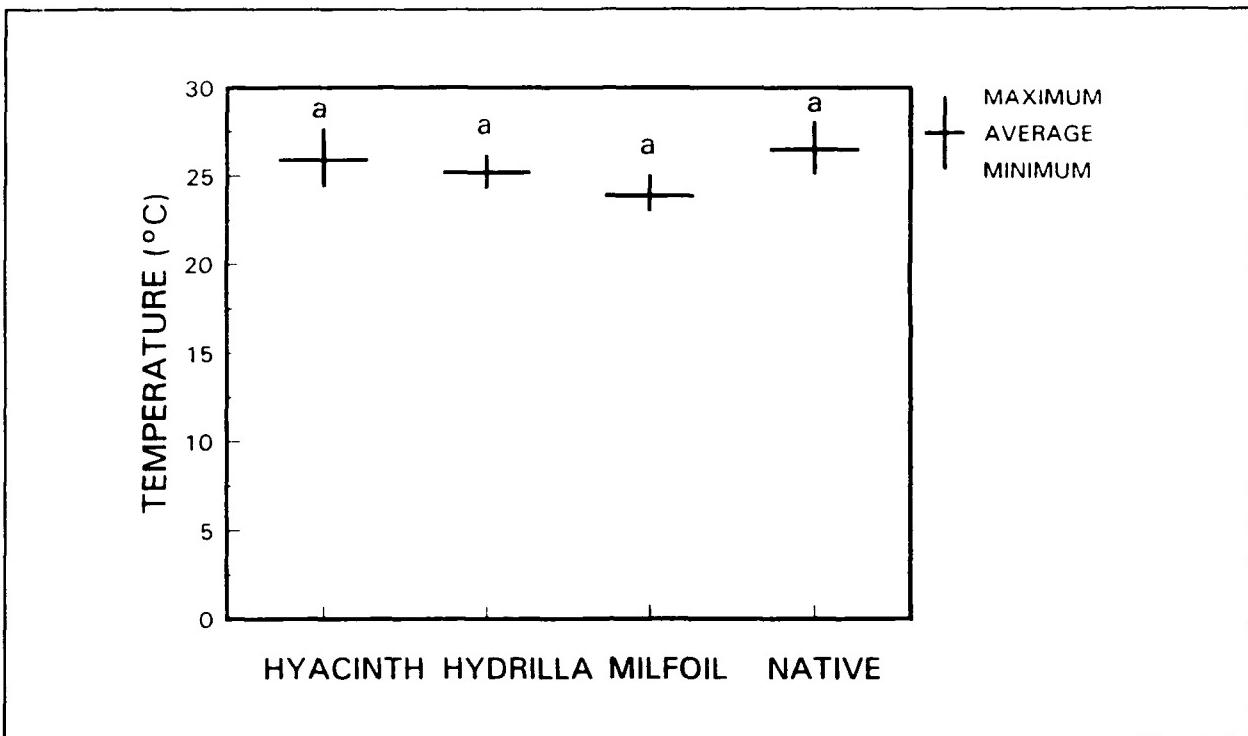


Figure 2. Means of daily averages, minima, and maxima for temperature (°C) for four ponds of species composition types. Means sharing the same letter do not differ significantly ($p=0.05$)

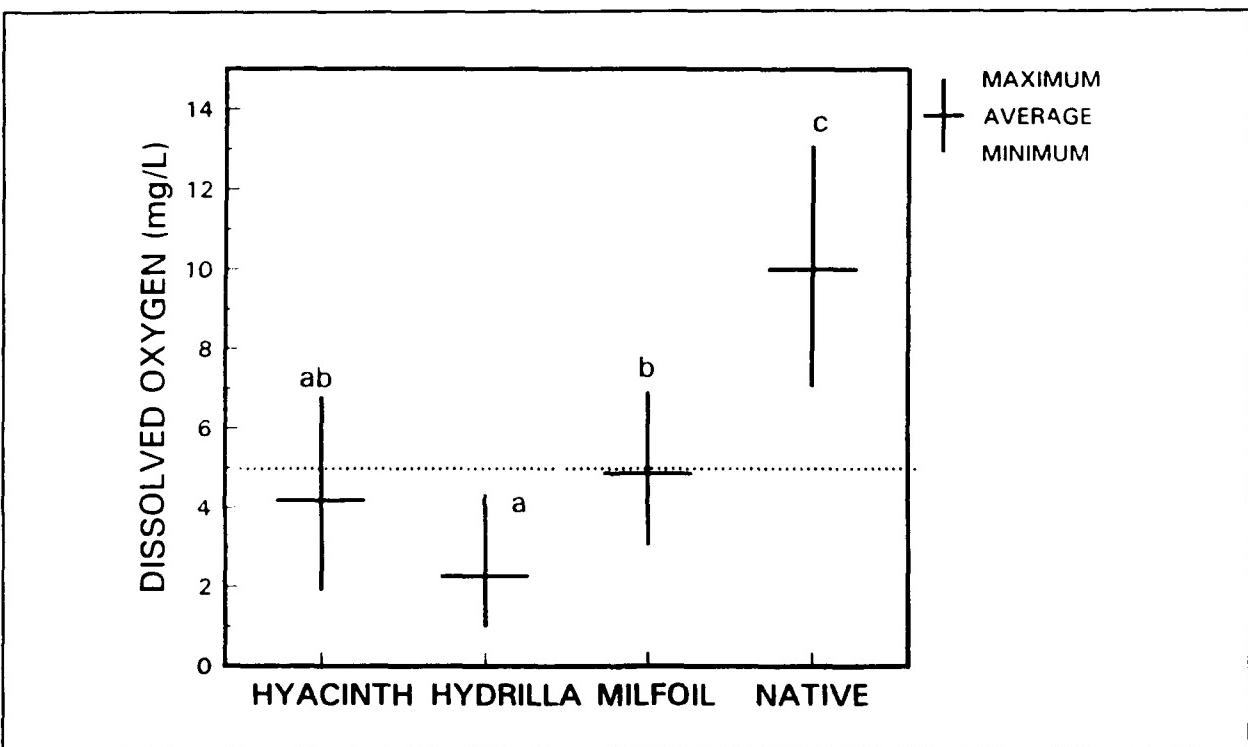


Figure 3. Means of daily averages, minima, and maxima for dissolved oxygen (mg/L) for ponds of four species composition types. Means with same letter do not differ significantly ($p=0.05$). The dotted line indicates the 5-mg/L undesirable level

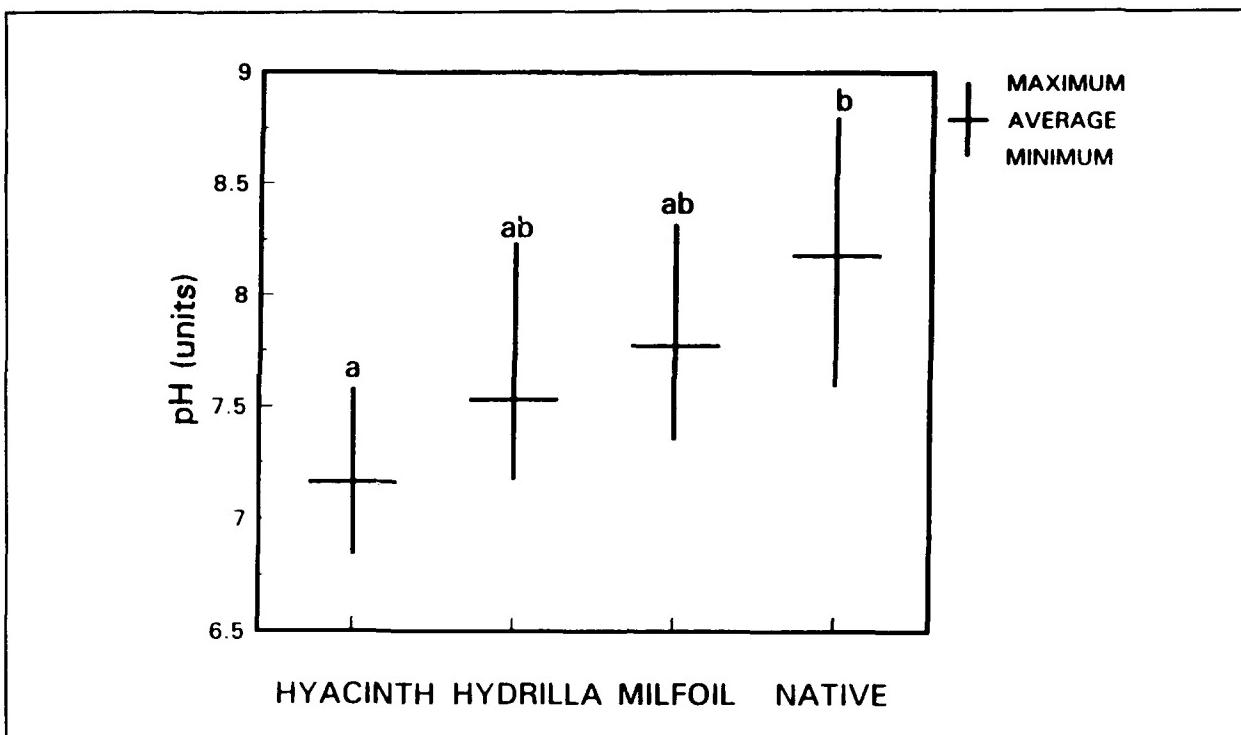


Figure 4. Means of daily averages, minima, and maxima for pH (units) for ponds of four species composition types. Means with same letter do not differ significantly ($p=0.05$)

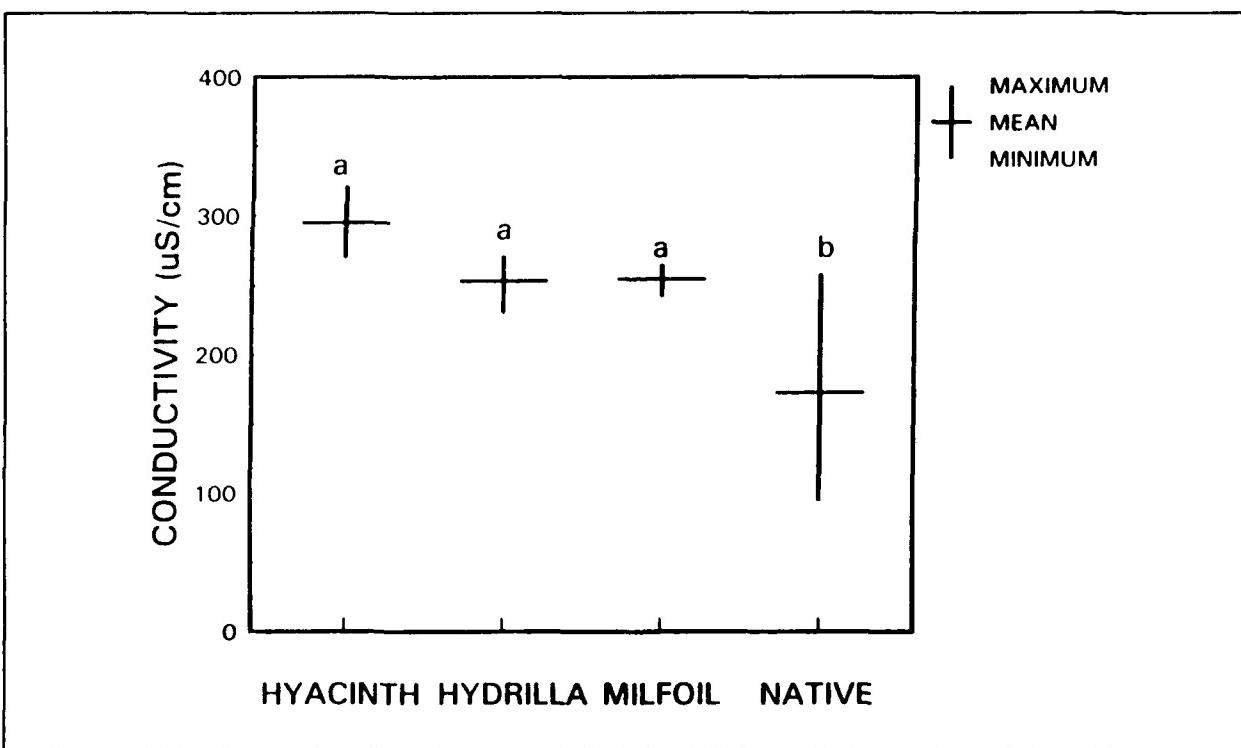


Figure 5. Means of daily averages, minima, and maxima for conductivity ($\mu\text{S}/\text{cm}$) for ponds of four species composition types. Means with same letter do not differ significantly ($p=0.05$)

Conclusions

Based on the results of this study, conclusions are summarized below.

- a. Vegetation had no significant effect on pond water temperature.
- b. Exotic species ponds showed significantly lower average DO values (generally lower than 5 mg/L) than did native species ponds. Native species ponds exhibited greater daily fluctuations, but averaged over 5 mg/L.
- c. Exotic species ponds generally exhibited higher pH values than did native species ponds.
- d. Native vegetation ponds exhibited greater daily fluctuations and significantly lower conductivity than did the exotic species ponds.

Ongoing Research

With the excellent opportunity afforded by the LAERF, we are continuing to monitor water quality in ponds of different species composition to expand data sets for full seasonal cycles. Additional parameters are also being analyzed on a routine basis by the on-site chemistry laboratory to develop a broader data set and a better understanding of the processes taking place in the ponds of varying aquatic plant communities.

Acknowledgment

Special thanks are expressed to Aleida Eu-banks, Susan Dutson, and Kristi Brandon for their assistance.

Environmental Characteristics of Ponds at the Lewisville Aquatic Ecosystem Research Facility

by

R. Michael Smart,¹ Joe R. Snow,¹ and Gary O. Dick¹

Introduction

The Lewisville Aquatic Ecosystem Research Facility (LAERF) includes 55 earthen ponds that are available for experimental research on aquatic and wetland ecosystems. In an ongoing effort to provide WES scientists and managers with needed information on the characteristics and utility of these experimental systems, we present preliminary information on the physical attributes and environmental characteristics (light, temperature, and water quality) of the ponds.

Physical Characteristics of Ponds

The ponds range in size from 0.4 to 1.9 acres (Table 1) and are of various shapes (Figure 1). Most of the ponds (37 of 55) are rectangular and range from 0.67 to 0.86 acre, making them suitable for conducting replicated experiments. Pond volumes range from 1 to 7 acre-ft, with most of the ponds holding between 2 and 3 acre-ft. Maximum pond depth ranges up to 9 ft, although average depth is much less, with most averaging about 3 ft in depth. This depth is suitable for conducting studies of both submersed and floating aquatic plant species as well as emergent wetland plants. In studies of submersed aquatic plants conducted at the LAERF to date, we have concentrated sampling in deeper portions of the ponds (Figure 2). However, the gentle slopes of the pond bottoms provide opportunities for sampling at various water depths if desired. Pond water levels are easily regulated, so wetland or other studies requiring shallower or variable water depths can be accommodated.

Meteorological Conditions

A meteorological station installed at the LAERF has been collecting data on solar radiation; wind speed and direction; temperatures of the air, soil, water, and sediment; and rainfall for nearly 2 years. Several ponds are equipped with data loggers and environmental monitoring equipment for obtaining continuous records of environmental conditions for specific studies. All of these data are available in digital form to researchers using the ponds.

Water temperatures measured in the ponds reflect seasonal changes in daily air temperature (Figure 3). Both water temperature and light intensity (Figure 4) exhibit strong seasonality and are representative of environmental conditions occurring over much of the continental United States.

Light intensity in the ponds decreases with increasing depth in a manner typical of clear lakes (Figure 5). The clarity of water provided to the ponds is high, allowing for high light levels and favorable growing conditions for submersed aquatic plants.

Water Quality

Water is provided to the ponds from Lewisville Lake, a water supply/flood control reservoir constructed and operated by the Corps of Engineers. The reservoir impounds the waters of the Elm Fork of the Trinity River and provides drinking water to the cities of Dallas and Denton, TX. Water supplied to the ponds is sampled and chemically analyzed on a regular

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Table 1
Physical Characteristics of LAERF Ponds

Pond No.	Surface Area, acres	Water Volume, acre-ft	Average Depth, ft	Area >2 ft Deep, %	Pond No.	Surface Area, acres	Water Volume, acre-ft	Average Depth, ft	Area >2 ft Deep, %
1	1.75	6.76	3.85	89	28	0.85	3.03	3.55	83
2	1.10	3.35	3.04	70	29	0.40	1.07	2.64	55
3	0.93	3.04	3.25	63	30	0.73	2.28	3.10	96
4	1.02	3.38	3.32	47	31	0.72	2.10	2.92	64
5	0.95	3.16	3.32	55	32	0.72	2.10	2.92	64
6	1.16	4.46	3.85	—	33	0.72	2.18	3.04	65
7	1.53	6.23	4.06	85	34	0.72	2.18	3.04	65
8	0.74	2.11	2.86	81	35	0.73	2.11	2.91	64
9	0.74	2.11	2.86	81	36	1.92	6.59	3.44	70
10	0.74	2.16	2.93	81	37	1.16	3.26	2.81	42
11	0.74	2.16	2.93	81	38	0.56	1.61	2.89	56
12	0.74	2.27	3.04	73	39	0.56	1.61	2.89	56
13	0.50	1.26	2.50	50	40	0.56	1.61	2.89	56
14	0.74	2.07	2.80	75	41	0.56	1.61	2.89	56
15	0.86	2.88	3.36	70	42	0.73	2.32	3.17	69
16	0.86	2.88	3.36	70	44	0.68	2.11	3.10	61
17	0.86	2.88	3.36	70	45	0.67	2.09	3.11	70
18	0.63	1.93	3.06	73	46	0.67	2.44	3.62	80
19	0.67	1.99	2.96	69	47	0.67	2.41	3.61	79
20	0.74	2.41	3.25	81	48	0.67	2.44	3.62	80
21	0.85	3.01	3.56	82	49	0.67	2.41	3.61	79
22	0.84	2.99	3.56	83	50	0.67	2.09	3.11	70
23	0.85	3.01	3.56	82	51	0.68	2.11	3.10	61
24	0.85	3.03	3.55	83	52	0.67	2.09	3.11	70
25	0.86	3.05	3.53	83	53	0.68	2.11	3.10	61
26	1.00	3.47	3.46	84	54	0.67	2.09	3.11	70
27	0.86	3.05	3.53	83	55	0.86	2.69	3.12	39

basis by LAERF personnel (Table 2). We also regularly monitor the composition of water leaving the facility. The chemical composition of many experimental and culture ponds is also monitored. These data, as well as additional onsite analytical services, are available to researchers using the ponds.

Alkalinity and pH of the water supply are generally stable throughout much of the growing season (Figure 6); however, aquatic plants cause dramatic changes in pond water chemistry. Example data shown here were obtained from a *Hydrilla* culture pond. The occurrence

of monospecific populations of different species of aquatic plants in ponds of initially similar water and sediment composition provides a unique opportunity to study the effects of aquatic plants on the physical and chemical characteristics of aquatic environments (see preceding paper).

Acknowledgment

Michael Crouch, Susan Dutson, Aleida Eubanks, David Honnell, John Madsen, Kimberly Mauermann, and Susan Monteleone of the LAERF contributed to this effort.

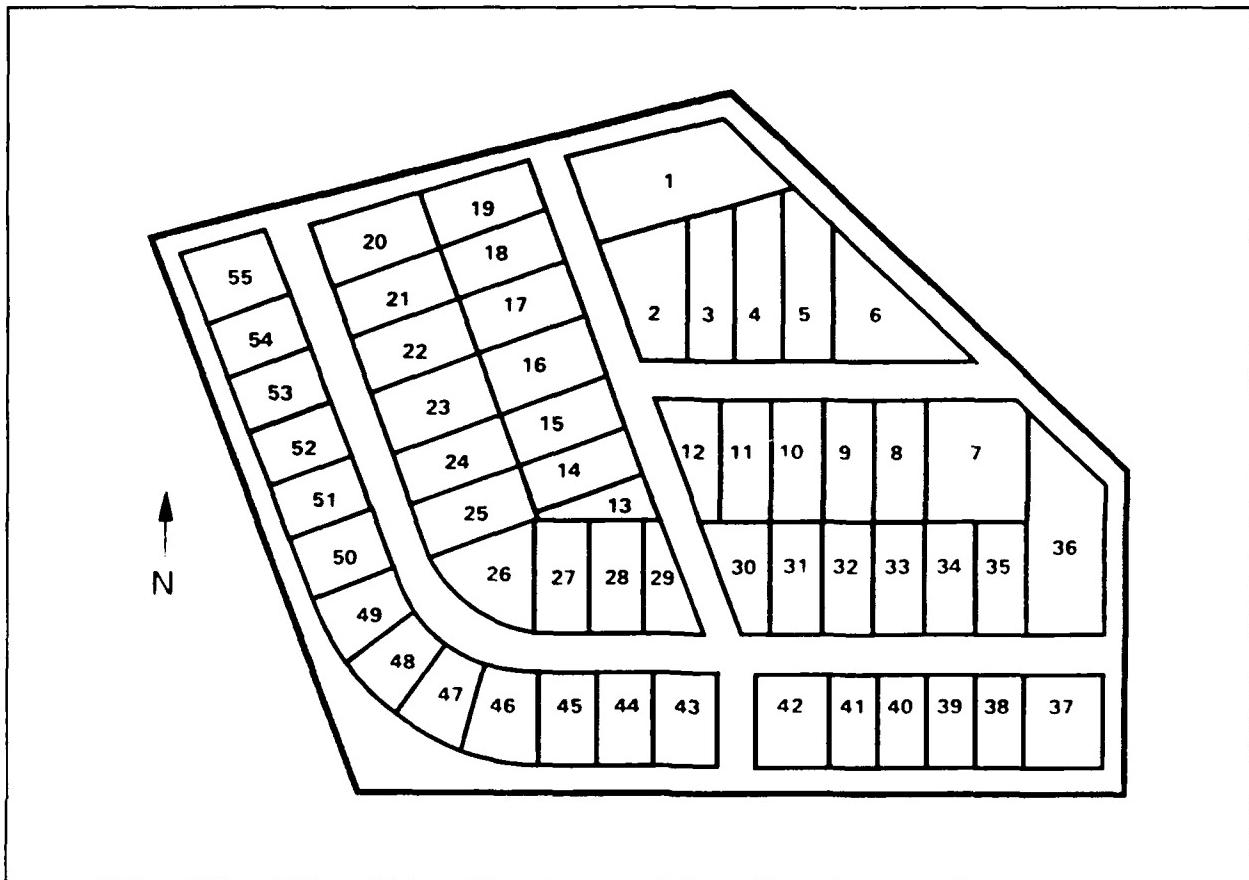


Figure 1. Configuration of LAERF ponds

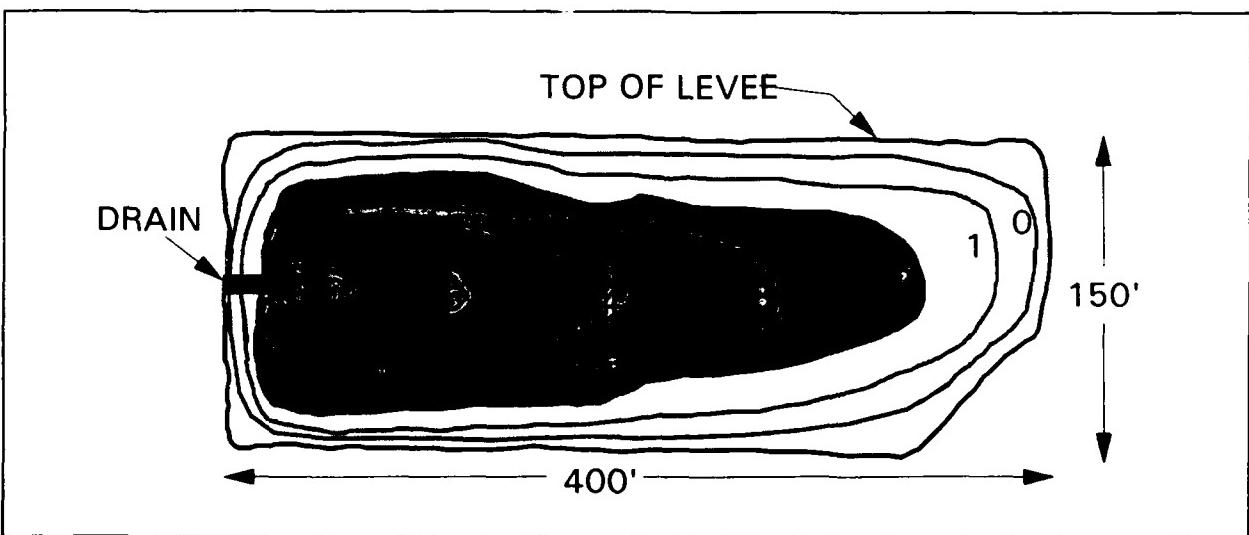


Figure 2. Depth contours of Pond 5 (in feet)

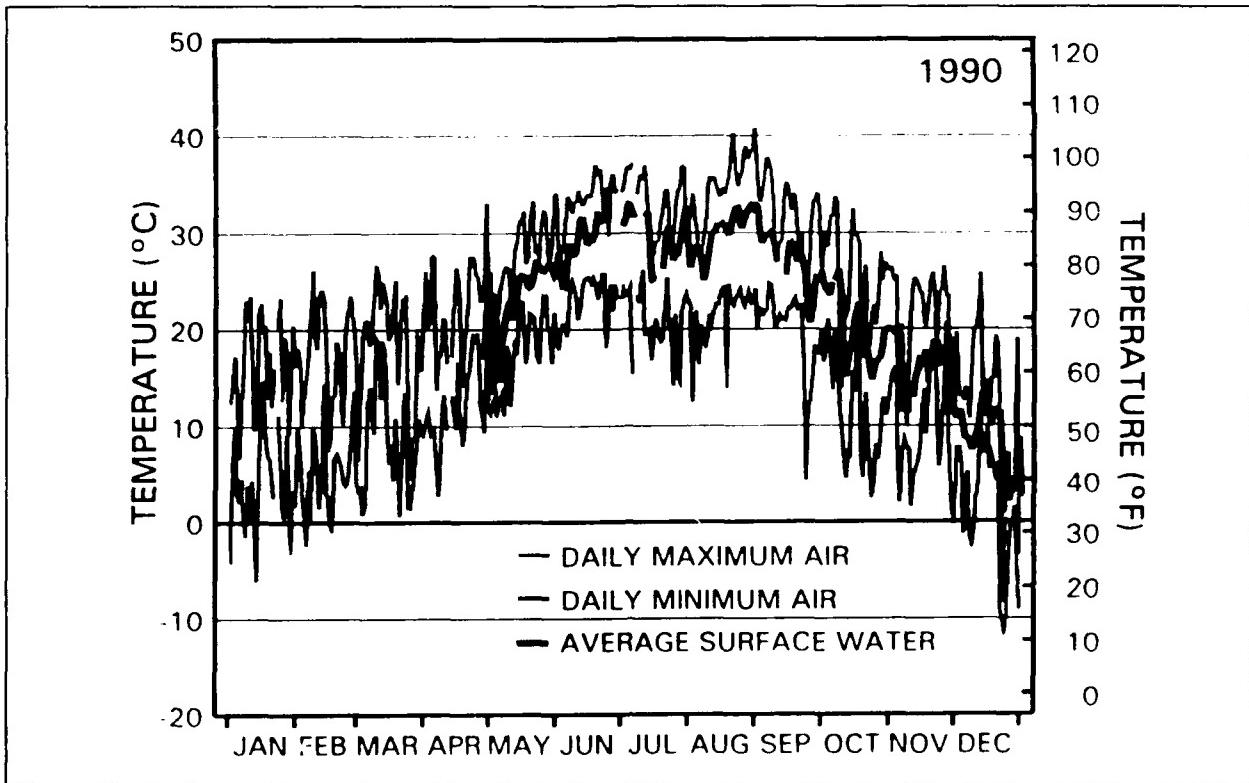


Figure 3. Daily minimum and maximum air and daily average surface water temperatures measured at the LAERF during 1990

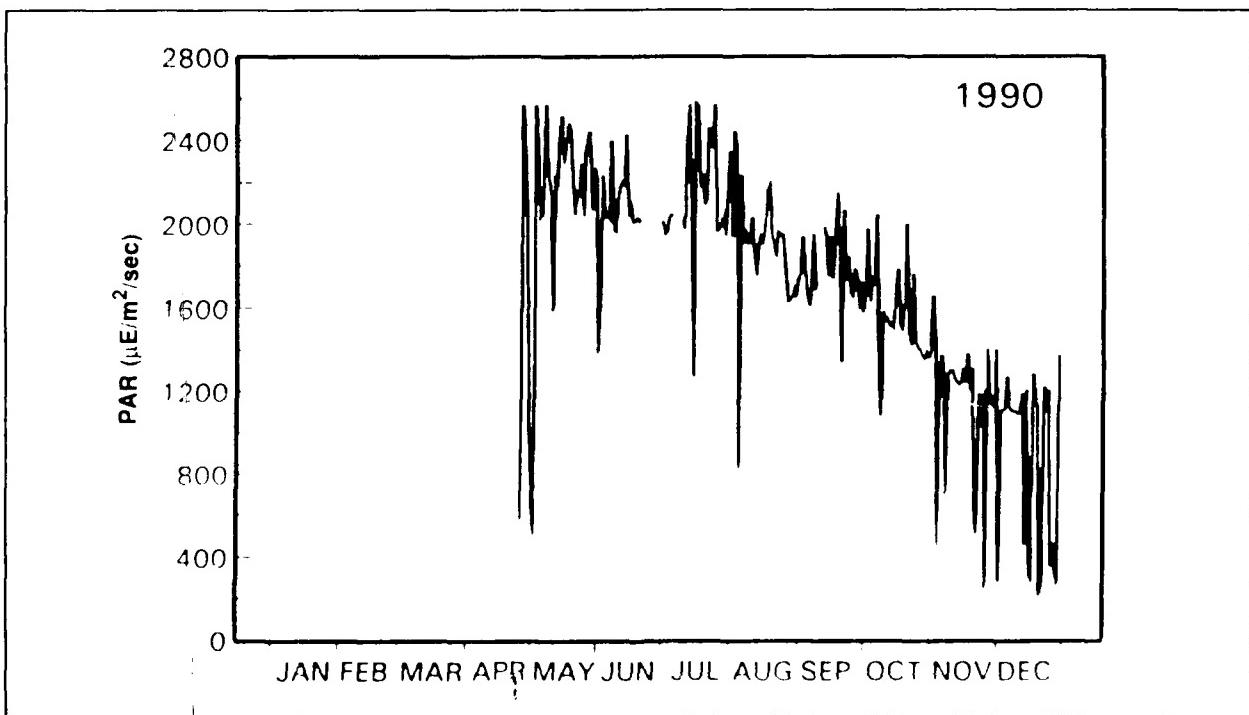


Figure 4. Light intensities (photosynthetically active radiation, PAR) measured at the LAERF during 1990

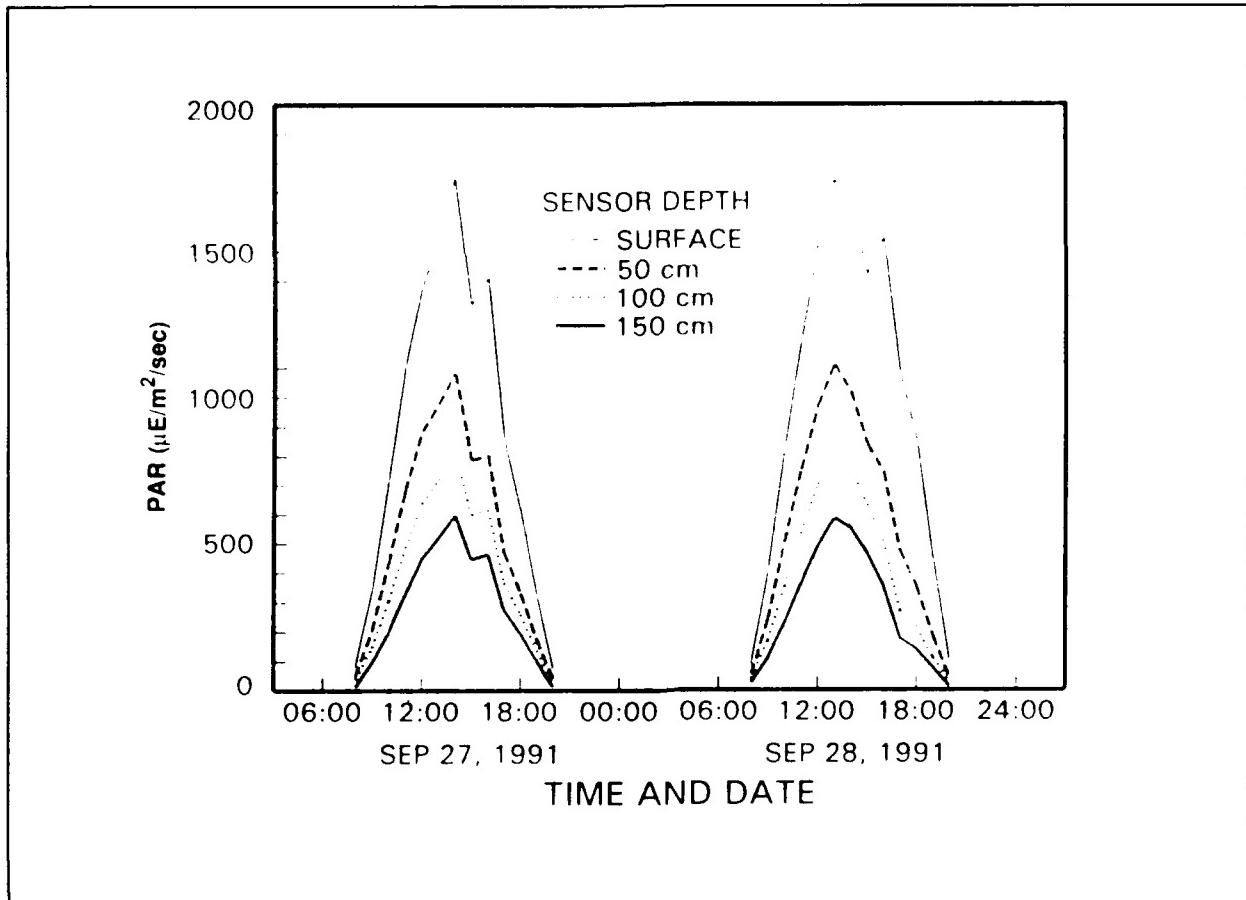


Figure 5. Light intensities (photosynthetically active radiation, PAR) over two diurnal cycles at the water surface and at different depths in Pond 26

Table 2 Water Quality Parameters Measured at LAERF	
Analytical Category	Parameters
Water quality	Temperature, pH, conductivity, dissolved oxygen, alkalinity, turbidity, total suspended solids, chlorophyll A
Nutrients	Nitrate nitrogen, ammonium nitrogen, soluble reactive phosphorus, total phosphorus
Metals	Sodium, potassium, calcium, magnesium, iron, manganese

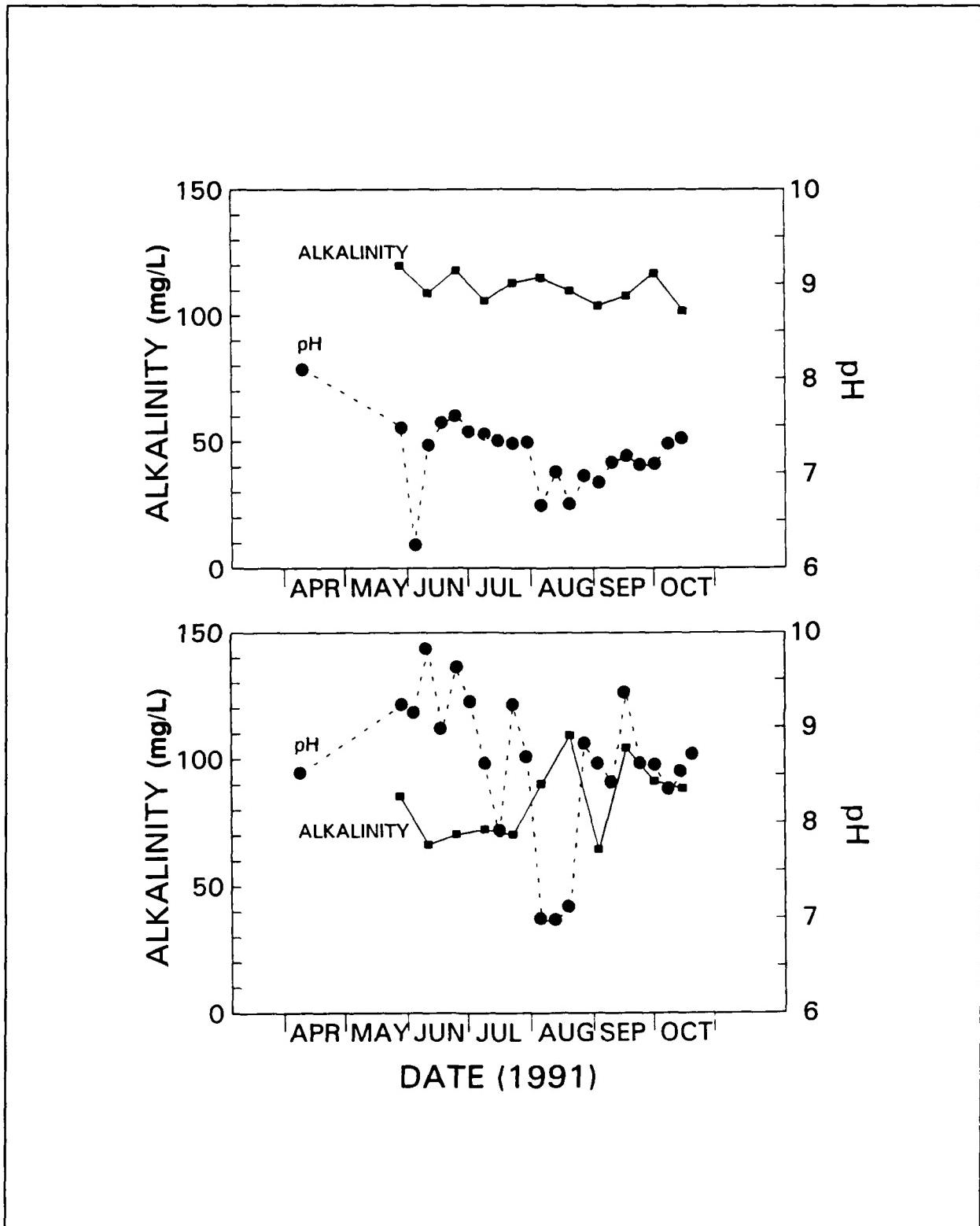


Figure 6. Alkalinity and pH of the LAERF water supply (upper) and in a *Hydrilla* culture pond (lower)

Annual Report - Aquatic Plant Control Operations Support Center

by

Wayne T. Jipsen¹

In October 1980, the Jacksonville District was designated by the Office, Chief of Engineers, as the Aquatic Plant Control Operations Support Center (APCOSC) in recognition of the District's knowledge and expertise gained through the administration of the largest and most diverse aquatic plant management program in the Corps. The APCOSC personnel assist other Corps elements and other Federal and State agencies in the planning and operational phases of aquatic plant control. The specific duties and relationships with other Corps APC programs, and guidelines for utilization of the APCOSC, are outlined in Engineer Regulation (ER) 1130-2-412.

The Center responded to 119 requests for assistance during fiscal year (FY) 1991. A breakdown of these activities appears in Table 1. Figure 1 indicates the types of information requested; Figure 2 provides a breakdown as to the source of information requests.

The demand for and type of services performed by the Center vary from year to year, based on the type of problems encountered by Corps elements and other agencies. Four basic types of information are requested: planning, operations, research, and training. Planning assistance includes determinations

of water body eligibility and allowable costs, computation for benefit-cost ratios, methods of data acquisition, and other factors that enter into the process of planning an Aquatic Plant Control Program. Operations assistance involves most aspects of chemical, mechanical, biological, and integrated technology. The Center provides data, information, and recommendations relating to operational activities. Information on research activities is provided to requestors if available, or the requests are referred to WES. Training assistance includes providing materials for use in educational and training programs, and presentation of the Pesticide Applicators Training Course and the Aquatic Plant Management Course by Center staff.

During FY 1991 the Center published the Information Exchange Bulletin, conducted an APC training course review for the Seminole Indian Tribe of Florida, collected and distributed alligatorweed flea beetles to a number of locations throughout the Southeast, and participated in a Congressional Office of Technology Assessment fact-finding visit. The Center also continued assisting OCE in the development of the APC Program Evaluation Guidance document and the revision of ER 1130-2-412.

¹ US Army Engineer District, Jacksonville; Jacksonville, FL..

Table 1
APCOSC-Support Assistance, FY 1991

Type Assistance	Corps				Other Federal	Foreign Country	State/Local	Industry	Private	Total
	OCE	WES	Division	District						
Planning	8	4	6	8	0	1	3	1	0	31
Operations	5	6	9	15	4	0	9	6	5	59
Research	1	10	0	2	3	0	5	0	0	21
Training	0	5	0	1	0	0	0	1	1	8
Total	14	25	15	26	7	1	17	8	6	119

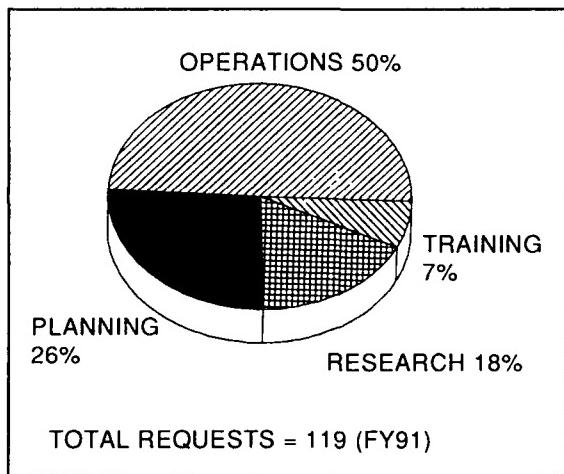


Figure 1. Types of information requested

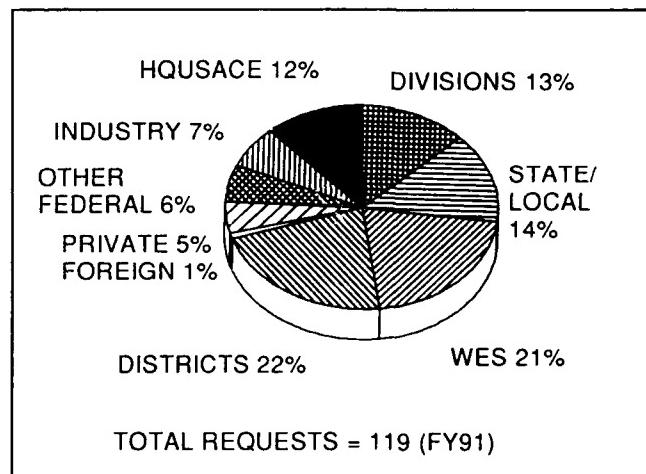


Figure 2. Sources of requests

Simulation Technology

Role of Simulation Technology in the Aquatic Plant Control Research Program and Status of Current Development Efforts

by

R. Michael Stewart¹

General

The purpose of this paper is to describe the role of Simulation Technology in the Aquatic Plant Control Research Program (APCRP) and to present an overview of current research activities. Development of simulation procedures was formally established as an APCRP technology area in fiscal year (FY) 1985. Since that time the technology area has expanded to include three research areas under which simulation procedures are being developed—Plant Growth Simulations (Work Unit 32440), Biocontrol Simulations (Work Unit 32438), and Chemical Control Simulations (Work Unit 32439). A fourth research area under Simulation Technology, Aquatic Plant Databases (Work Unit 32506), was established to provide digital environmental database technology support for development, testing, and execution of the various simulation procedures.

Role in the APCRP

The activities undertaken during the development of each simulation procedure can be categorized into three distinct roles or functions in the APCRP: conceptual design and model formulation, testing and evaluation, and technology transfer. These functions are illustrated in Figure 1, and brief descriptions of these functional areas are provided in the following sections.

Conceptual design and model formulation

During development of all simulation procedures, the first function is the analysis of information on the "system" being modeled. Work efforts that are completed during this phase of the development process include the review of pertinent literature, development of a conceptual model or framework of the system, and development of a first-generation computer-based simulation procedure. Each of these activities is discussed briefly below.

Literature review. An extensive literature review is logically the first step undertaken in the development of any simulation procedure. It is through this process that all pertinent information is collected and analyzed in order to determine if the "knowledge base" is sufficient to model a given process or system. The information gathered and analyzed through the literature review is then used to facilitate development of the conceptual framework for the simulation procedure.

Conceptual design. The conceptual design provides a functional organization and representation of system processes to be modeled. Because of the nature and complexity of the systems to be modeled, information gaps are often identified during development of the conceptual framework. For some of these gaps, assumptions are generated to allow completion of the conceptual framework.

¹ US Army Engineer Waterways Experiment Station, Vicksburg, MS.

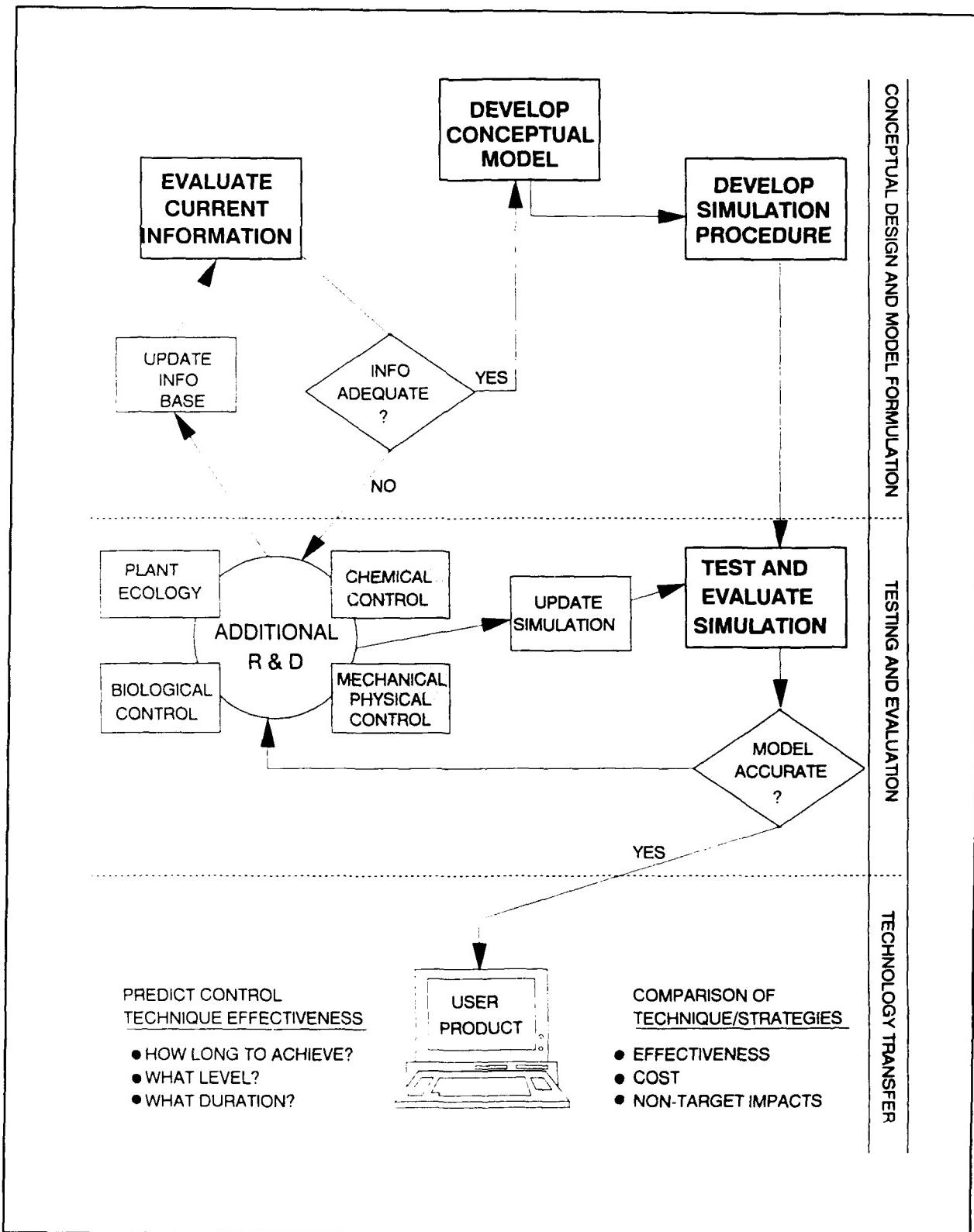


Figure 1. Functional categorization of work activities during development of APCRP simulation procedures

However, in some instances, research efforts must be undertaken to provide the needed information and relationships before the conceptual design can be completely formulated.

Development of first-generation model. The final step toward synthesizing the information is the development of a first-generation simulation procedure. For this purpose, mathematical equations and computer-based algorithms are developed to simulate various processes included in the conceptual design. Digital databases of environmental variables must be developed to support execution and generation of model outputs. Finally, user interface routines are developed to facilitate model execution and interpretation of simulation outputs.

Testing and evaluation

The first-generation code developed represents our current knowledge base related to the system being modeled. Testing and evaluation of the first-generation code includes step-by-step analysis of individual processes and the interactions between various processes. As needed, the model evaluation process includes acquisition of new information and development of new relationships to improve our knowledge base of the system being modeled.

First-generation code. Model testing and evaluation are undertaken to ensure the functionality and accuracy of the relationships incorporated in the first-generation code. During this stage, the simulation procedure is evaluated by comparing model outputs with ground truth data. This allows testing and evaluation of the conceptual design of the total predictive system as well as the accuracy of the mathematical relationships used to represent the individual system components or processes. Assumptions incorporated in the conceptual design are especially scrutinized.

Updates and further testing. Key research and development needs that should be collaboratively undertaken by the various APCRP Technology areas (i.e. Plant Ecology, Chemical Control, Biocontrol) are often de-

tected during this step. The simulation model and model results are especially useful in determining and pointing to key research needs by providing a procedure for systematically evaluating interactions among aquatic plants, control agents, and the environmental conditions. As new information becomes available through collaborative APCRP research, the simulation procedures are updated with improved relationships, and the evaluation step is continued with additional testing.

Technology transfer

The computer-based simulation procedures provide two levels of technology transfer. The first level of transfer accomplishes exchange of information between APCRP technology areas. This level of technology transfer begins during development of the conceptual model and continues through the evaluation process. The second level helps accomplish the transfer of information from APCRP technology areas to end-users such as reservoir managers. This level of transfer is accomplished through distribution of the model and user documentation. Training sessions are also conducted as necessary for effective use/understanding of the simulation packages.

Overview of Existing Development Efforts

Plant growth simulations

The objective of this research is to develop personal computer (PC) based plant growth models for certain aquatic plant species that will generate simulation outputs needed to evaluate how growth of these plant species will respond to site-specific environmental conditions. These simulation procedures will assist in predicting levels of aquatic plant infestations and will serve as a basis for development of simulation procedures of control techniques developed under other APCRP technology areas.

Currently, first-generation simulation procedures have been developed for waterhyacinth, hydrilla, and Eurasian watermilfoil. The first-generation waterhyacinth model has

been completed and will be released during FY92 through distribution of the **INSECT** Version 1.0 software package. Research activities are continuing to provide needed improvements to the waterhyacinth simulation procedure. These research activities include small-scale pond studies and laboratory investigations (Madsen 1992) of seasonal changes in respiration rates, daughter plant production, and seasonal biomass partitioning. Research efforts are also under way for collection of field data needed for evaluation of the **HYDRILLA** and **MILFOIL** plant growth simulations (Wooten and Stewart 1991). During FY92, these field data collection efforts will continue at Guntersville Reservoir, Alabama.

Additionally, validation studies conducted to date indicate that current relationships included in spring regrowth algorithms need improvement. Small-scale pond studies and tank studies will be conducted at the Lewisville facility to provide data needed to make these improvements. These studies focus on biomass partitioning during regrowth and the vertical growth potential of different over-wintering structures of hydrilla and Eurasian watermilfoil. These Lewisville studies are coordinated closely with ongoing research in the APCRP Plant Phenology research area.

Biological control simulations

The objective of this research is to develop simulation procedures for currently available biological control agents of certain nuisance aquatic plants. These procedures couple simulations of the biological control agent's population dynamics with a plant growth simulation of the targeted aquatic plant species. Outputs from these simulation procedures provide answers to "what if" questions related to the effects of site-specific environmental conditions on the population dynamics of the biological control agent, and subsequently, what level of control the biological control agent will exert on the target plant infestation.

Currently, simulation capabilities exist for the biocontrol of waterhyacinth by two wee-

vil species (**INSECT** Version 1.0) and of several submersed plant species by white amur fish (**AMUR STOCK** Version 1.5). Applications of these existing simulation procedures are described in Boyd and Stewart (1990, 1991, and 1992). A User's Manual for the **INSECT** simulation procedure will be published during FY92. Additionally, a new version of the **INSECT** simulation is under development to allow consideration of hydrilla control by a fly species, *Hydrellia pakistanae*. Boyd and Stewart (1992) provide a status summary on this research effort.

Chemical control simulations

The research includes development of simulation procedures for the chemical control of nuisance aquatic plant species. These procedures consider the effects of site-specific conditions on the postapplication fate of aquatic herbicides, including partitioning in sediments, water, and plant tissues. From these time-dependent concentration estimates, the simulations predict/estimate the level of control effected by the herbicide application.

Chemical control simulation capabilities have been developed for several herbicide formulations and target plant combinations. Recent status summaries and example applications can be found in Rodgers, Clifford, and Stewart (1991) and Stewart (1992).

Final documentation for the current version of this simulation procedure, **HERBICIDE** Version 1.0, will be available in FY92. During FY91 and continuing into FY92, we are jointly participating in the construction of a mesocosm test facility for herbicide fate and effects studies at the Lewisville facility. Additionally, through this work effort, we are supporting research conducted under the Herbicide Delivery System work unit (32437) to obtain better understanding of herbicide loading within plant tissues and plant mortality.

Aquatic plant databases

The objective of this research is to develop procedures and digital environmental databases that are compatible with computer-based

simulation procedures developed under other Simulation Technology task areas. The resulting databases will be structured to support initialization of the simulation procedures and to provide a method for storing, visualizing, and analyzing simulation outputs.

Recent efforts have focused on the development of digital databases that are compatible with PC-based Geographic Information System (GIS) packages. These efforts are described in Kress, Causey, and Ballard (1990) and Welch and Remillard (1991). Ongoing database development efforts utilizing GIS techniques are described by Kress and Causey (1992).

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Status and Application of WES AMUR/STOCK and INSECT Models

by

William A. Boyd¹ and R. Michael Stewart¹

Introduction

The US Army Engineer Waterways Experiment Station (WES) is developing personal computer-based software packages to assist in the understanding of, and transfer of information relevant to, aquatic plant control technologies developed by WES under the Aquatic Plant Control Research Program. Currently at WES there exist two biological control simulations, the White Amur Stocking Rate (AMUR/STOCK Version 1.5) simulation and the Insect (INSECT Version 1.0) simulation. Neither AMUR/STOCK nor INSECT has reached its final form; however, the interim simulation procedures will be released to selected researchers and managers for their evaluation. These evaluations as well as ongoing research will allow future technical improvements to these biocontrol simulation procedures.

AMUR/STOCK (Version 1.5)

Status

The original AMUR/STOCK simulation was developed to provide information useful for determining proper stocking rates of diploid white amur fish for control of hydrilla in water bodies having user-specified site characteristics. Miller and Decell (1984) provide guidance for proper use of AMUR/STOCK Version 1.0.

In Version 1.5 of AMUR/STOCK, two additional simulation capabilities have been added. These provide outputs for seasonal growth (i.e. biomass) of Eurasian watermilfoil and an annual aquatic plant species complex found in Guntersville Reservoir as affected

by water temperature, photoperiod, season, water body carrying capacity, and white amur consumption. Improvements were also made to the fish feeding and growth relationships used in AMUR/STOCK, to allow simulations to consider each of the three white amur genetic variants: diploids, triploids, and hybrids.

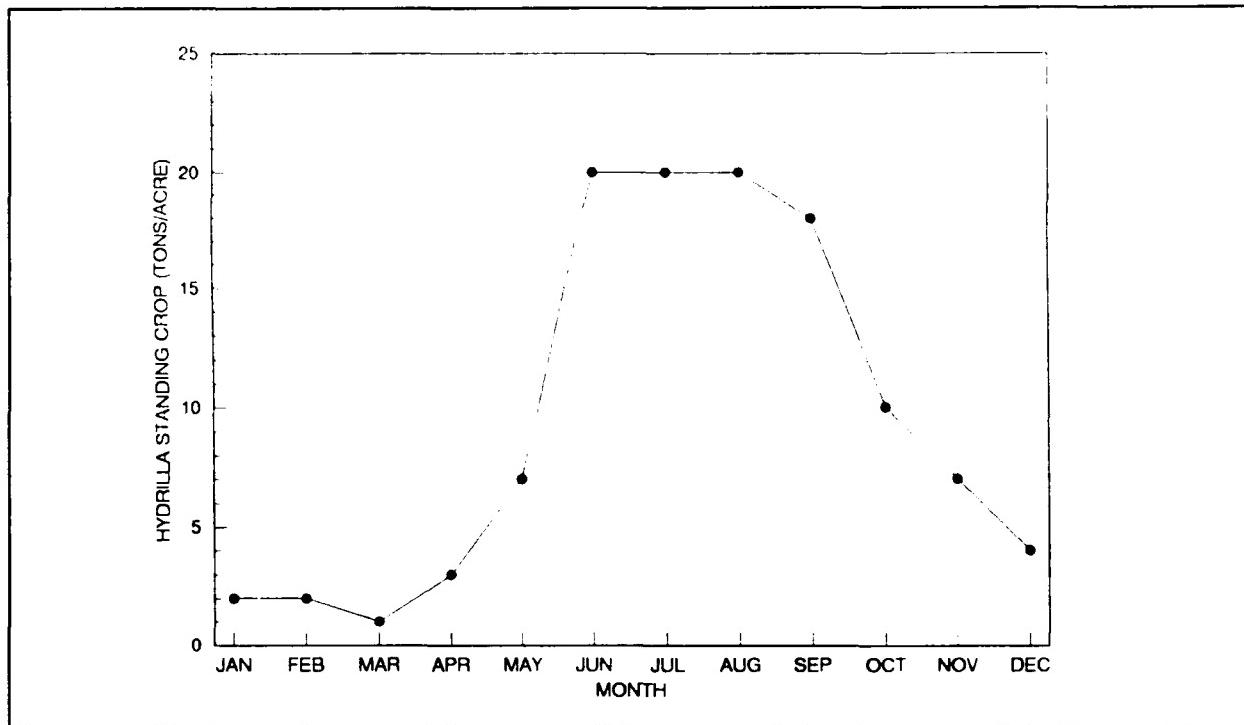
In general, these improvements incorporate white amur feeding, growth, and mortality relationships included in the Illinois Herbivorous Fish Simulation System as reported in Wiley et al. (1985) and Wiley and Wike (1986). Modifications to these relationships were made during comparison of initial AMUR/STOCK simulation output with data reported for the white amur demonstration project in Guntersville Reservoir (Tennessee Valley Authority (TVA) 1987, 1989).

Applications

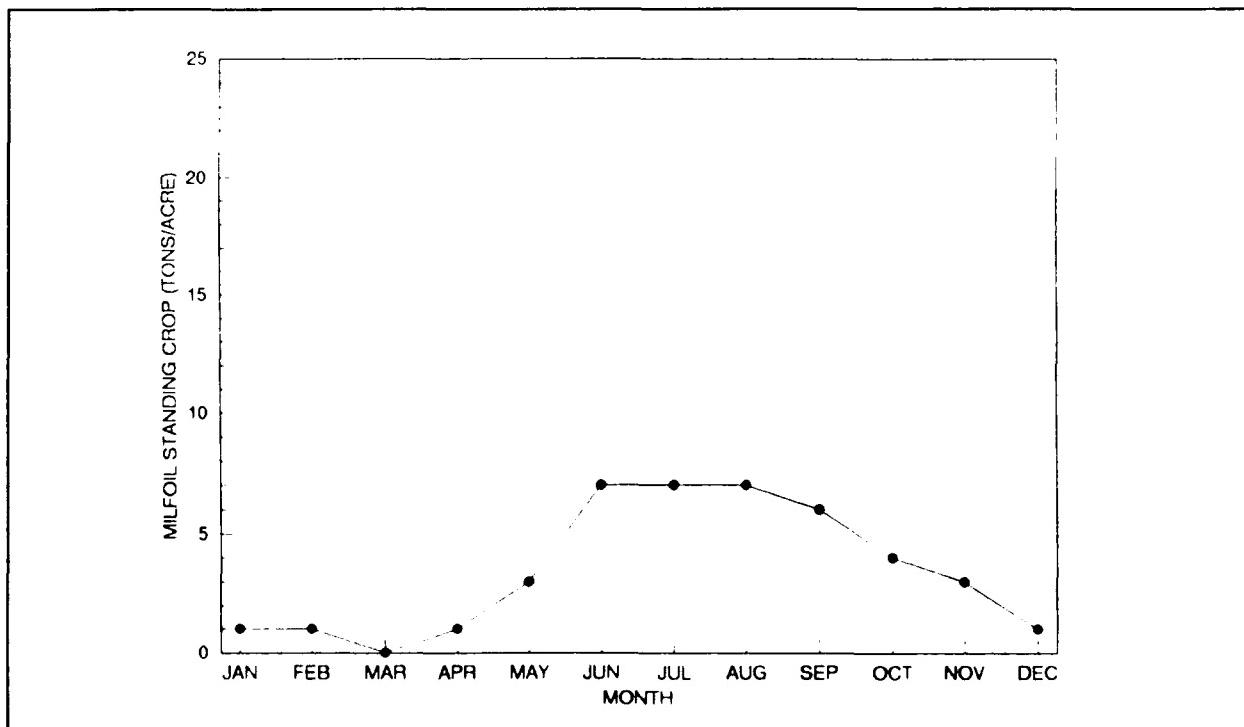
AMUR/STOCK was developed to provide users with a systematic evaluation tool for answering "what if" questions regarding the results of proposed white amur stocking scenarios for managing nuisance growth of aquatic plants. These scenarios could consider general conditions for any water body, or site-specific conditions and specific stocking plans. The various scenarios under question may also differ by a number of other factors (Boyd and Stewart 1990).

In a more general context, AMUR/STOCK can be used to determine the level of control achieved by stockings of specified numbers and sizes of white amur under a defined set of environmental conditions and plant growth patterns. Figures 1-3 show "hypothetical" seasonal plant growth patterns for each of

¹ US Army Engineer Waterways Experiment Station, Vicksburg, MS.



*Figure 1. Seasonal plant growth pattern for hydrilla
without impacts from fish feeding*



*Figure 2. Seasonal plant growth pattern for Eurasian watermilfoil
without impacts from fish feeding*

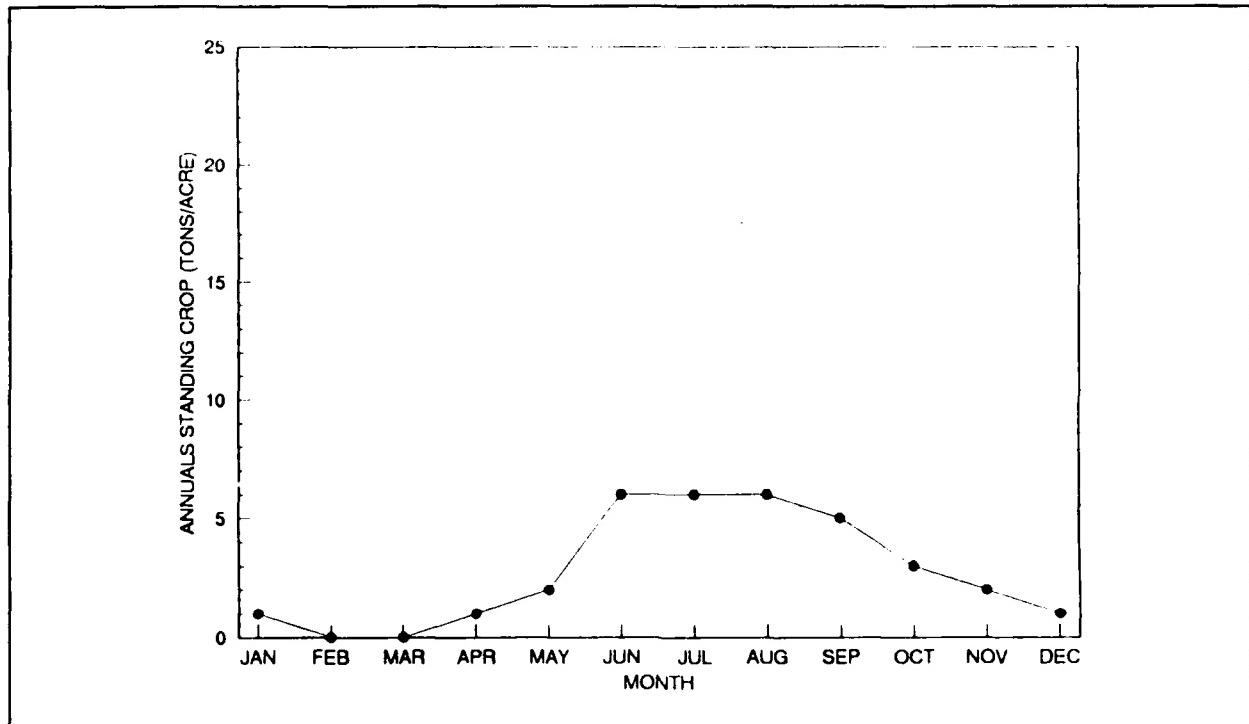


Figure 3. Seasonal plant growth pattern for the annuals without impacts from fish feeding

three plant species without impacts from fish feeding. Figure 4 presents the control predicted as a result of stocking 50,000 triploid white amur, weighing 0.75 lb each, in Year 1 of a 10-year simulation period for each of three plant species. Considering the stocking of 50,000 fish as described, Figure 4 indicates that maximum control would be achieved for each plant species during Year 6 of the 10-year simulation period, where 50,000 triploids were shown to control 9,695 acres of the annuals, 3,549 acres of Eurasian watermilfoil, and 1,907 acres of hydrilla. Based on this information and the size of the area in which complete control is desired, the number of fish needed to provide the desired level of control could be determined.

To illustrate a "site-specific" application, simulation outputs were generated for white amur stocking scenarios developed for 1990 Guntersville Reservoir environmental conditions and stocking plans proposed by the TVA. Model initialization conditions include the following: (a) pretreatment infested acreage by plant species based on aerial surveys con-

ducted by TVA during the fall of 1989, (b) estimates of maximum seasonal standing crop by plant species based on biomass samples collected at Guntersville Reservoir during 1990, (c) number and size of white amur stocked by TVA during 1990, and (d) actual dates (i.e. month) of stocking by TVA. These data were used for model initialization conditions and are shown in Table 1.

The proportions of fish used in the separate simulations for each plant species are shown in Table 2. These proportions are based on the set of initialization values for 1990 Guntersville Reservoir conditions (see Table 1) as well as plant preference and availability assumptions considered by Boyd and Stewart (1991).

Infested acreages remaining for each plant type are shown in Table 3. Values presented are infested acreage amounts prior to white amur stocking (i.e., YR0) and poststocking infested acreage amounts for a 10-year period (i.e., YR1-YR10) calculated from AMUR/STOCK simulation outputs. Based on assumed fish proportions for the three plant

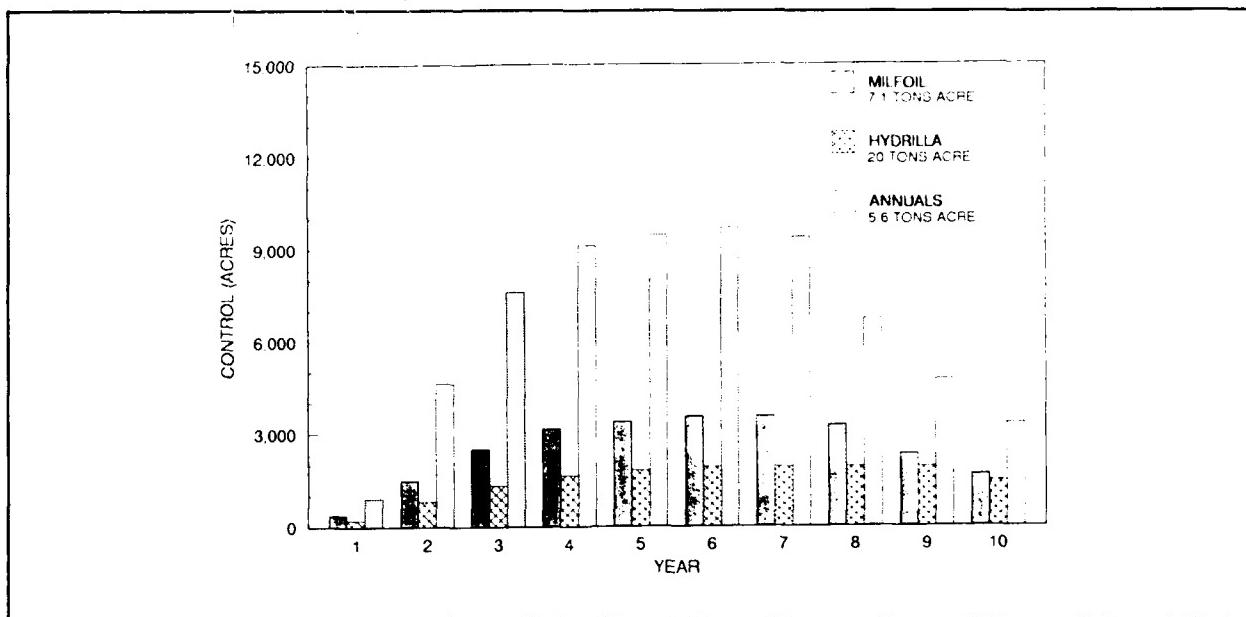


Figure 4. Control achieved as a result of stocking 50,000 triploid white amur, stocked in April of Year 1, weighing 0.75 lb each

Table 1
Simulation Conditions for 1990

Plant Species	Infested Area, acreage	Maximum Seasonal Biomass, tons/acre
Eurasian watermilfoil	7,000	7.1
Hydrilla	2,000	20.0
Annual species	1,000	5.6
White Amur Stocked		
Stocking Time	Number Stocked	Average Size, lb
May, Year 1	35,000	0.75
June, Year 1	50,000	0.75
July, Year 1	15,000	0.75

Table 2
Assumed Proportions of Fish Feeding on Each Plant Type During Each Year of Simulations for 1990 Conditions

Plant Type	YR1	YR2	YR3	YR4	YR5	YR6	YR7	YR8	YR9	YR10
Annuals	0.50	0.11	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
Hydrilla	0.50	0.89	0.78	0.60	0.55	0.52	0.52	0.52	0.52	0.67
Milfoil	0.00	0.00	0.22	0.40	0.45	0.48	0.48	0.48	0.48	0.33

Table 3
Infested Acreage Remaining for Each Plant Type

Plant Type	Prestocking YR0	Poststocking									
		YR1	YR2	YR3	YR4	YR5	YR6	YR7	YR8	YR9	YR10
Annuals	1,000	736	0	0	0	0	0	0	0	0	0
Hydrilla	2,000	1,736	768	0	0	0	0	0	0	0	0
Milfoil	7,000	7,000	7,000	5,990	4,382	3,859	3,538	3,538	3,730	4,637	5,832

species and prestocking infested acreage amounts, the model outputs show complete control of the annuals by the end of Year 1 and of hydrilla by the end of Year 2. By Year 3, with the annuals and hydrilla controlled, the model outputs show reduction in the infested acreage of Eurasian watermilfoil. During succeeding years, the milfoil acreage is reduced further until Year 8 when, due to fish mortality, the acreage begins to increase.

Goals for FY92

Goals for FY92 include preparation of needed documentation for AMUR/STOCK Version 1.5, including both a User's Manual and a Technical Report. As mentioned in Boyd and Stewart (1990) and Boyd and Stewart (1991), assumptions were made as to how the fish would feed based upon fish-feeding preference and availability of the plant types. Since many of the preferred plant types are widely dispersed throughout Guntersville Reservoir, fish travel patterns should be clarified to verify the assumptions made for fish feeding. Additionally, biomass data will continue to be collected for specified sites at Guntersville Reservoir. These data will then be used for comparison with predicted values and for refinement of plant growth and re-growth relationships.

INSECT (Version 1.0)

Status

INSECT is a biological control simulation procedure that includes interactions between a plant growth module for waterhyacinth and a biocontrol agent module for two species of *Neochetina* weevils. The INSECT simulation procedure is described in detail by Akbay, Wooten, and Howell (1988) and updates to the simulation are given by Howell, Akbay, and Stewart (1988) and Howell and Stewart (1989). The simulation (INSECT Version 1.0) is being released to selected researchers and managers along with an Interim User's Manual (Stewart and Boyd, in preparation). Additional technical improvements will be made to the INSECT simulation as ongoing research

clarifies important relationships considered within this biocontrol system.

Applications

The INSECT simulation procedure was developed to provide information needed to systematically evaluate how environmental conditions affect the growth and development of waterhyacinth, the population dynamics and life cycle processes of the *Neochetina* populations, and the plant/herbivore interactions that drive the overall biological control system. The ultimate goal of INSECT, as an information tool, is to assist in decision-making related to operational control of waterhyacinth. Though this goal has not been achieved thus far, INSECT is a very beneficial research and development tool for insect biocontrol efforts seeking to identify and better understand the relationships that govern this complex biological system.

Table 4 provides a list of the INSECT output parameters for both waterhyacinth and *Neochetina*. Figures 5 and 6 present a typical application of INSECT. Simulated output for numbers of *Neochetina* adults and larvae (per square meter) is shown plotted against actual field data through a 360-day simulation period. Ground truth data are shown with 95-percent confidence intervals of the means. While the simulation and field data appear to match well for *Neochetina* adults (Figure 5), there are significant differences between the simulation and field data for the larvae (Figure 6). The simulation output shows peaks at several times during the year where the field data do not. INSECT allows the researcher to isolate

Table 4
INSECT Output Parameters

Waterhyacinth		
Biomass	Plant Loss (Detritus)	
<i>Neochetina</i>		
Number of Plants	Plant Loss (Insect)	
Gross Production	Net Change in Biomass	
Respiration Cost Losses		
<i>Neochetina</i>		
Eggs	Pupae	
Larvae (1st, 2nd, & 3rd instar)	Adults	

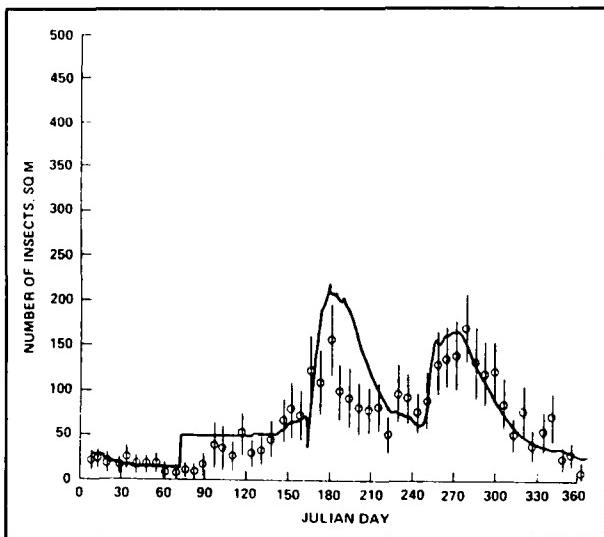


Figure 5. Example of INSECT simulation output for adult *Neochetina* plotted against field data with 95-percent confidence intervals of the means

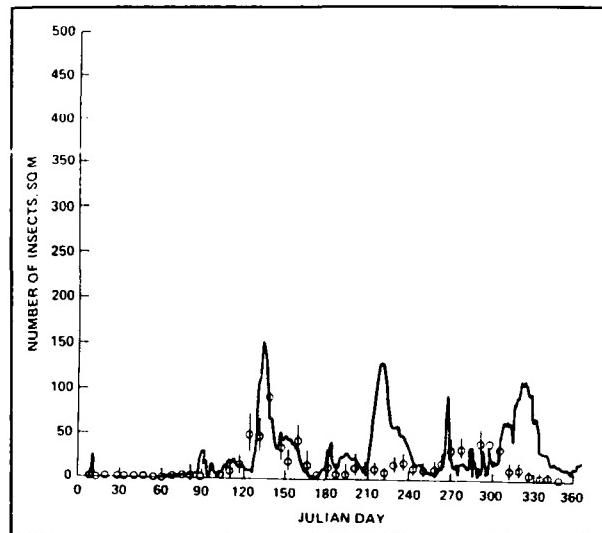


Figure 6. Example of INSECT simulation output for *Neochetina* larvae plotted against field data with 95-percent confidence intervals of the means

relationships used in the simulation procedure that may contribute significantly to differences between simulation output and field data.

Fecundity is one such relationship used by INSECT that is suspected of needing further elucidation. It represents the number of viable eggs produced by the adult *Neochetina*, and consequently has a profound effect on subsequent generations. In INSECT, fecundity is a function of temperature. The nutritional profile of waterhyacinth is also believed to exert great influence on the *Neochetina* weevils' reproductive condition (see Grodowitz and Freedman 1990). Thus, any significant shift in the nutrient level of the plants could either increase or decrease the number of ovulated eggs produced by adult weevils. Another important relationship is emigration, the number of adults that actually leave the site being simulated. INSECT assumes that emigration is a function of the number of adult weevils per plant.

A better understanding of these and other relationships, as well as how these are affected by certain environmental conditions, is vital. After these relationships have been isolated using INSECT simulations and substantiated

through research, they can be incorporated into the simulation procedure in order to produce results that more closely emulate what actually happens in the field.

Simulation is currently under development which will include interactions between hydrilla (*Hydrilla verticillata*) and the *Hydrellia pakistanae* fly. At present, a development time module for *Hydrellia pakistanae* has been developed for incorporation into this overall simulation capability. This module is driven solely by temperature and allows consideration of factors such as the number of generations and the time of development over a period of time.

Figure 7 illustrates duration of successive generations of *Hydrellia pakistanae* as a result of the accumulation of required degree-days over the course of a calendar year. Preliminary studies indicate a relatively short period of insect development time for *Hydrellia pakistanae*. Parametrized values used in this module are shown in Table 5. These values are based on the results of an initial study conducted by WES (see Warren 1992) and consider the impact of temperature on *Hydrellia pakistanae*.

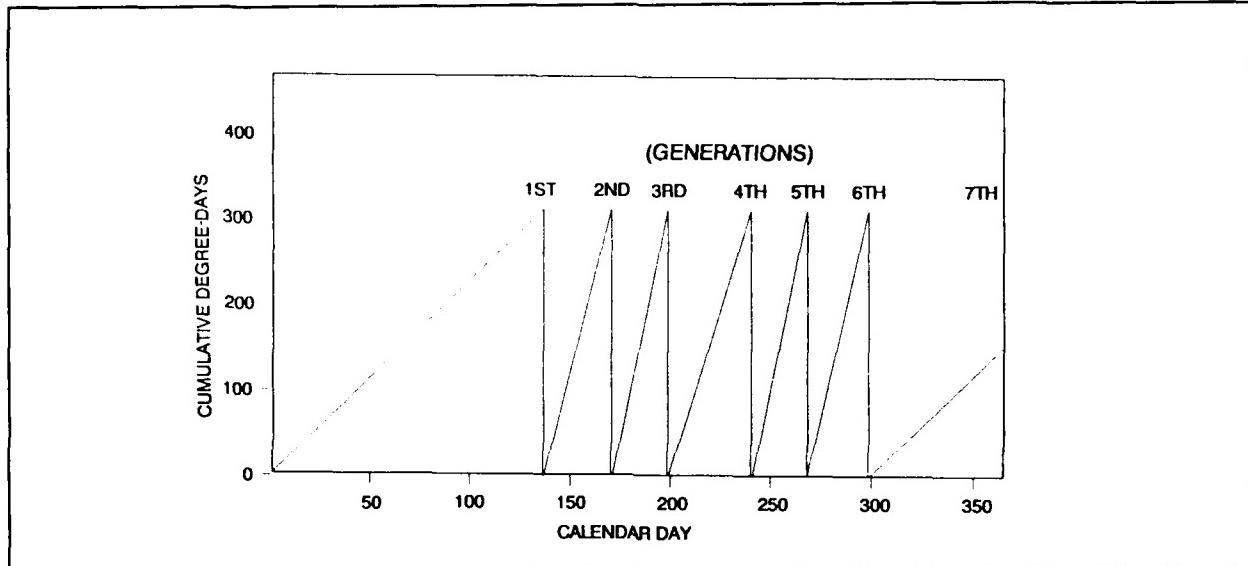


Figure 7. Development time of successive *Hydrellia pakistanae* generations (egg to adult) as a function of cumulative degree-days

Table 5 Development Time of <i>Hydrellia pakistanae</i>		
Life Stage	Threshold Temperature for Development, °C	Required Degree-Days
Egg	11	27
Larvae		
1st	11	54
2nd	11	51
3rd	11	76
Pupae	11	100
TOTAL		308

Goals for FY92

Improvements are planned for INSECT Version 1.0 during 1992. Updates will be based on user feedback as a result of releasing INSECT to users and from in-house testing, evaluation, and application. Also, additional development work is planned for the *Hydrellia pakistanae* (INSECT Version 2.0) simulation procedure.

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HERBICIDE, the Aquatic Herbicide Fate and Target Plant Effects Simulation Model

by

R. Michael Stewart¹

Description of HERBICIDE

The HERBICIDE simulation model is a decision support software package designed to produce output data useful for designing effective aquatic herbicide application strategies. The generalized structure of the HERBICIDE model (Figure 1) includes three modules that interactively estimate or predict the post-

application fate of the active ingredient of the herbicide formulation, the effectiveness of the herbicide treatment on the target plant infestation, and the posteffect response or regrowth of the target plant. The status of HERBICIDE was most recently presented in Rodgers, Clifford, and Stewart (1991), and the following paragraphs describe the function of each of the modules.

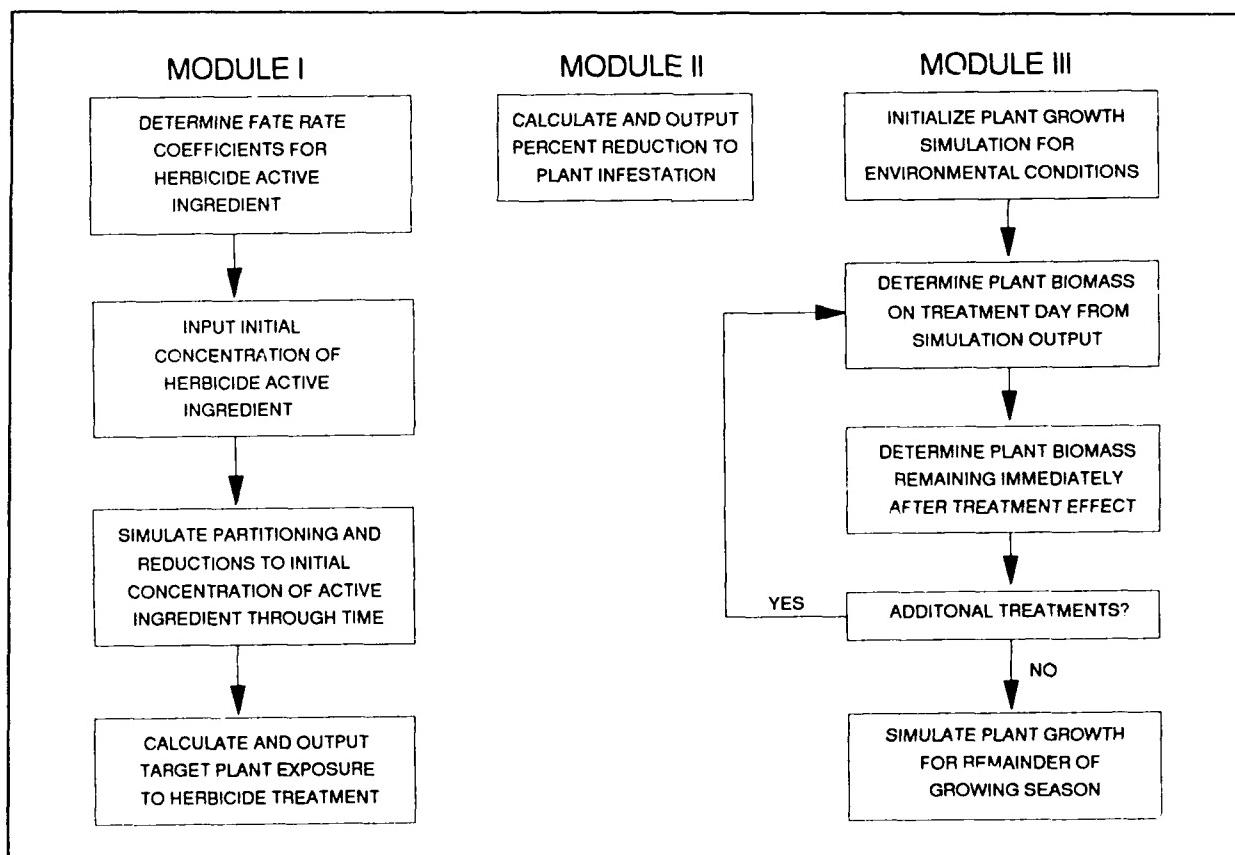


Figure 1. Generalized structure of the HERBICIDE simulation model

¹ US Army Engineer Waterways Experiment Station, Vicksburg, MS.

Herbicide Fate module

Various types of fate processes effect significant reductions to initial concentrations of aquatic herbicide active ingredients following their application or release into aquatic systems. These fate processes, working collectively, produce time-varying levels of the active ingredients within different "partitions" of an aquatic system. Aquatic system partitions considered by HERBICIDE are the sediments, the tissues of the target plant, and the water.

Module I of the HERBICIDE model allows consideration of the effects of major fate processes on herbicide formulation active ingredients. The effects that various fate processes have are dependent upon both site conditions and the properties of the aquatic herbicide formulation (Reinert and Rodgers 1987; Westerdahl and Getsinger 1988). Failure to consider the effects that major transfer and transformation processes have on aquatic herbicides when designing aquatic herbicide applications often results in attainment of lower levels of control than desired. The HERBICIDE model provides time-dependent simulation outputs for concentrations of the herbicide active ingredients within the different "partitions" in consideration of the effects of the major fate processes over time. Rate coefficients for the various fate processes are calculated based on user response to the input requirements shown in Table 1.

Herbicide Treatment Effectiveness module

The simulation outputs from this module provide information directly related to herbicide concentration/exposure time relationships being developed for plant mortality by APCRP Chemical Control Technology research reported by Netherland (1991) and Netherland, Green, and Getsinger (1991). As these referenced and other studies have shown, the level of control (i.e., plant mortality) achieved can be estimated from the level of exposure (i.e., concentration \times time) of the target plant to the herbicide. These studies also indicate that these exposure/mortality relationships

**Table 1
Input Requirements of Module I
for Calibration of Various Herbicide Fate
Process Algorithms in HERBICIDE**

Transfer Processes	Input Requirements
Drift	Percent loss of active ingredient
Dilution	Application rate of formulation Percent active ingredient fraction Release half-life of formulation Average depth of treated area
Sorption	Herbicide sediment layer partition coefficient Total suspended solids Sedimentation rate Depth of active sediment layer Sediment water content (percent) Sediment diffusion exchange rate
Volatilization	Volatilization half-life in water
Bioaccumulation	Bioaccumulation of active ingredient
Transformation Processes	Input Requirements
Oxidation	Oxidation half-life in water Oxidation half-life in sediments
Hydrolysis	Hydrolysis half-life in water Hydrolysis half-life in sediments
Photolysis	Photolysis half-life in water Photolysis half-life in sediments
Biodegradation	Biodegradation half-life in water Biodegradation half-life in sediments

Source: Rodgers, Clifford, and Stewart (1988).

are unique for each combination of herbicide active ingredient and target plant.

Module II of HERBICIDE includes mathematical equations that can be calibrated for different herbicide exposure/plant mortality relationships. The main output from this module is an estimate of the level of mortality (i.e., percent kill) inflicted on the target plant infestation.

Target Plant Response module

The ability of an aquatic plant infestation to respond to a herbicide treatment is the product of many factors, including the type of damage inflicted by the herbicide treatment (i.e., complete plant kill versus shoot injury); the level of damage inflicted; the response potential or growth rate of the plant species; the

phenological state of the plant (see Luu and Pesacreta 1988); site conditions; season; and others. The combination of these factors determines how a targeted plant infestation will respond to a herbicide treatment.

The function of Module III of HERBICIDE is to consider these factors and provide simulation output that estimates the regrowth response of a target plant infestation to a herbicide treatment. Module III will consist of functional versions of plant growth simulations developed for waterhyacinth, Eurasian watermilfoil, and hydrilla under the Simulation Technology work unit (32440). Initialization of Module III simulations will be based on outputs from Module II, model-supplied "default" data, and user inputs.

Status of HERBICIDE

HERBICIDE (Version 1.0) has been completed and produces fate (Module I) and effects (Module II) simulation outputs for the herbicide and target plant combinations listed in Table 2. The model outputs for 2,4-D (DMA) applications to waterhyacinth have been tested with data from both large- and small-scale field studies (Clifford, Rodgers, and Stewart 1990). Fate coefficients and effects relationships for the additional herbicide and target plant combinations included in Table 2 have been calibrated using available literature.

Table 2
Aquatic Herbicide Formulations and Target Plant Combinations Considered in Modules I and II of HERBICIDE (Version 1.0)

Target Plant	Herbicide Formulation
Waterhyacinth	2,4-D (DMA) Diquat
Eurasian watermilfoil	2,4-D (BEE) Diquat Endothall (Aquathol) Fluridone (Sonar SRP)
Hydrilla	Diquat Endothall (Aquathol K)

Documentation and software release of HERBICIDE (Version 1.0) will be accomplished during fiscal year 1992. User feedback resulting from the software and supporting documentation release will be used to improve usability of this decision support tool.

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Studies for Further Development of Existing WES Plant Growth Models

by
John D. Madsen¹

Introduction

Simulation modeling is an important research activity toward improving our understanding of aquatic plants and their management. Once developed, simulation procedures fulfill three distinct needs: (1) prediction of trends, (2) elucidation of gaps in our knowledge, and (3) analysis of the relative weights of causes and effects in the regulation of plant populations.

The ability to predict trends in plant populations is the application most commonly associated with simulation procedures. The ultimate end product is a model that can predict plant growth patterns for specified site conditions, thereby providing information needed to design management strategies. However, this is not the only area in which simulations serve a useful function. In the process of developing these procedures, gaps in both data (raw information) and knowledge (understanding of underlying mechanisms or principles) are exposed. In this manner, simulation modeling can bring together scientists from different technology areas to work together on solving problems. In addition, simulation procedures can provide a procedure for evaluating interactions of plant population dynamics and underlying mechanisms to determine the effectiveness of control techniques.

As part of a continuing effort to develop existing WES simulations, waterhyacinth (*Eichhornia crassipes*) growth and development was studied at the Lewisville Aquatic Ecosystem Research Facility. Physiological parameters investigated included leaf photosynthesis and respiration, root and stembase

respiration, leaf turnover, and daughter plant turnover. Information reported herein, and data currently being collected, will be used to evaluate and improve plant growth relationships currently included in existing models.

Methods and Materials

Waterhyacinth studies were conducted in two ponds: a reference pond to which no fertilizer was added, and a nitrogen-enriched pond to which 11.4 kg of ammonium sulfate was added weekly. In addition, hay and hydrochloric acid were added to both ponds to maintain pH at or below 8; Aquashade was also added to a concentration of 1 ppm to reduce algal growth. Environmental data were recorded by an Omnidata datalogging system at 5-min intervals. Parameters measured included air and water temperatures, incident light (PAR), total incoming solar radiation, and humidity.

Leaf photosynthesis and respiration

Leaf photosynthesis data were collected using an ADC portable photosynthesis system of an open design that measures carbon dioxide changes between a reference path and a leaf chamber by infrared gas analysis. The leaf chamber was a specially designed 49-ml Plexiglas clamp, which also measured leaf temperature. Leaf photosynthesis measurements were taken and recorded for six waterhyacinth leaves in each of the two ponds. Then, each leaf was placed in an environment exclusive of light; respiration measurements were taken after an approximate 15-min acclimation period to these "darkened" conditions.

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All measurements were taken after exchange rates had stabilized. Leaf photosynthesis and respiration measurements were taken approximately every 2 weeks from July through October 1991.

Root and stembase respiration

Waterhyacinth root and stembase dark respiration rates were examined for roots and stembases separated from healthy plants. After separation, the roots and stembases were placed in darkened 300-ml biological oxygen demand (BOD) bottles with pond water and incubated for periods of 4 to 24 hr, depending on the temperature of incubation. Respiration was determined by measuring oxygen consumption over the incubation time period using a YSI model 58 oxygen meter and BOD bottle probe, corrected for water-borne oxygen consumption using darkened blank bottles. Incubation temperatures were maintained using a Remcor circulating heater/chiller, with temperatures maintained to $\pm 1^{\circ}\text{C}$.

Roots and stembases were examined from plants of both the reference and nitrogen-added pond populations. Respiration was measured at ambient temperature approximately every 2 weeks beginning in late May. In addition, respiration response across temperature was examined at three time periods, when ambient temperatures were low, medium, and high. Data for the low temperature-acclimated plants are presented herein; other data are being used for further analysis.

Leaf and daughter turnover

The production and loss of both individual leaves and daughter plants were examined using a leaf and daughter tagging method. These experiments were performed in 0.25-m² wire rings placed in both the reference and nitrogen ponds. Into each ring, one rosette was placed at the initial time period, and all leaves and daughter plants were tagged using small cable ties.

Each week, these plants were reexamined, with all tagged and untagged leaves and

daughter plants counted. Untagged leaves and daughter plants were then tagged. Leaf death was determined when the lamina had turned completely brown and/or had fallen off. Daughter loss was classified as when the stolon connecting the daughter and parent plant was severed. Generally, the daughter plant was still viable at this point, and capable of self-sufficiency.

Six rings were used for each cohort in each of the two ponds, with three cohorts examined: early (late May), mid-season (early July), and late-season (mid-September). These plants were monitored until senescence was complete; data presented are from 1990, as data collection has not been completed for the 1991 growing season. Air temperature data collected at the pond were used to relate production and loss rates to ambient environmental conditions. For regressions, raw data points of individuals are used, but the points plotted are means for a given week and cohort to reduce graphic clutter.

Results and Discussion

In the experiments reported herein, the following two questions were being addressed: (1) Does the fertility level affect the particular plant physiological mechanisms, and (2) Is there a relationship between the plant physiological mechanism and a given environmental factor, such as temperature or light intensity?

Leaf photosynthesis and respiration

Although both photosynthetic and respiratory rates of leaves varied seasonally, no obvious patterns related to nutrient level, light intensity, or temperature were noted (Figure 1). Additional data are needed to further clarify photosynthetic relationships to temperature and light. However, no significant difference was observed in either respiration or photosynthesis of leaves between reference and nitrogen-added populations.

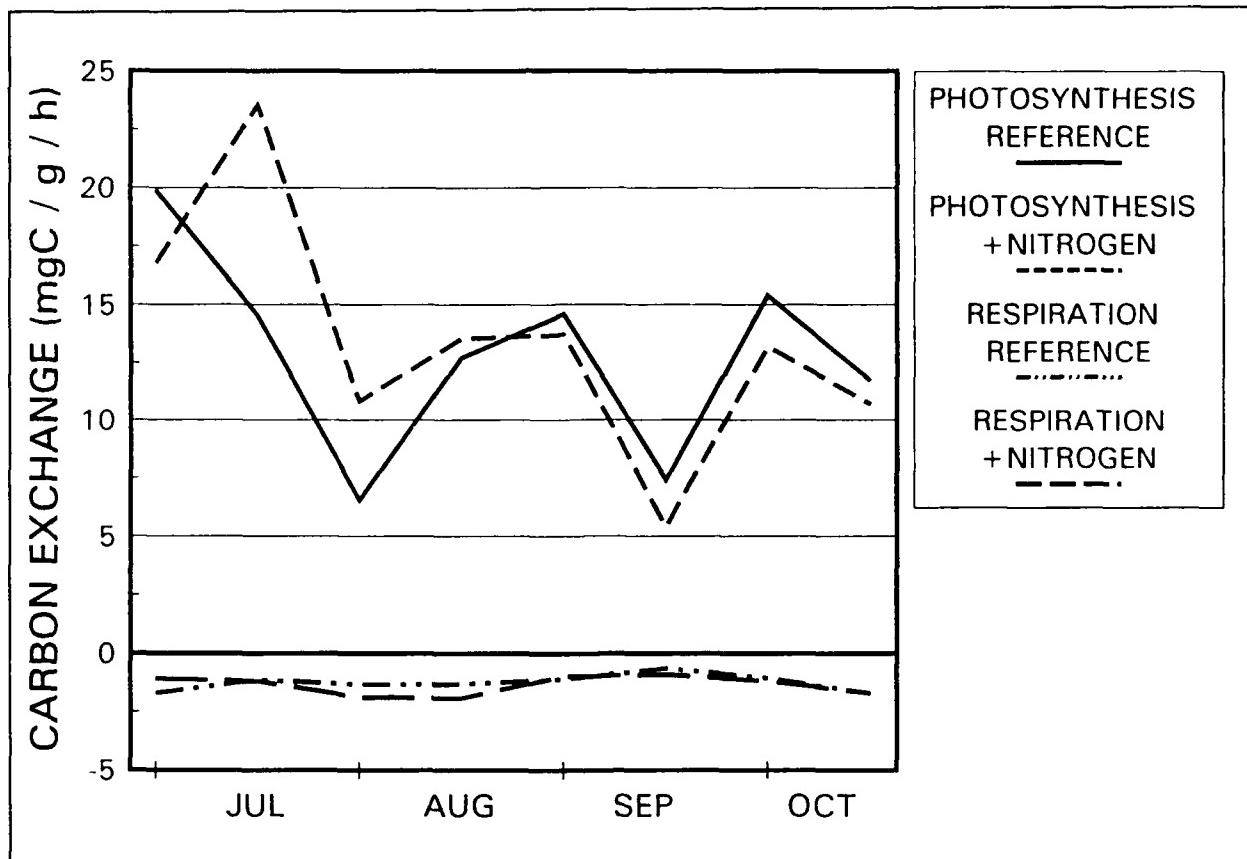


Figure 1. Waterhyacinth leaf photosynthesis and respiration for populations from reference and nitrogen ponds across the growing season as measured by carbon exchange ($\text{mg C g}^{-1} \text{ hr}^{-1}$)

Root and stembase respiration

A much larger database is currently available for dark respiration of roots and stembases. In Figure 2, the relationship of respiration to temperature is shown for roots and stembases that were preconditioned at a low ambient temperature. From these data, several points are evident. As with leaf photosynthesis and respiration, there was no significant difference between populations from the reference pond and nitrogen pond. However, different tissues (e.g., root versus stembase) exhibit markedly different respiration rates, with root tissues having a respiratory rate up to three times higher than stembases. This observation is supported by the relatively active role of roots in the uptake of nutrients as opposed to the predominantly passive storage role of stembases. Models involving respiration rate must account for these differences,

as well as incorporating plant part allocation patterns.

Third, temperature had a significant effect on respiration rate, particularly at higher temperatures. Ambient temperature was a good estimator of respiration rate. Finally, unpublished data indicate that the ambient preconditioning temperature also altered the resultant respiration rate.

Leaf and daughter turnover

In Figure 3, data on leaf gain (A) and loss (B) are shown for all cohorts in both reference and nitrogen ponds as a function of average weekly temperature for the sample week. As with the leaf photosynthesis and respiration rates discussed previously, no significant effect of nutritional treatment was observed. Air temperature was correlated to

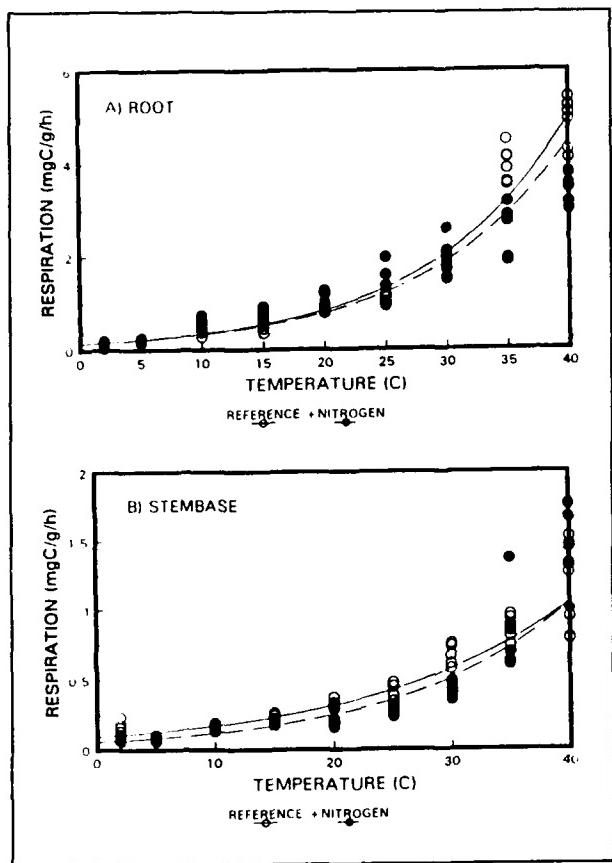


Figure 2. Respiration rates ($\text{mg C g}^{-1} \text{ hr}^{-1}$) for roots (A) and stembases (B) of waterhyacinth plants from reference and nitrogen ponds at a low ambient temperature tested across a range of temperatures from 2 to 40 °C

leaf production, with an average of two leaves produced every week at 30 °C. Leaf loss, however, did not appear to be correlated to temperature, with an average of one leaf per week lost throughout the growing season. The one caveat to this statement is that sub-freezing temperatures, which frequently occur during early spring and late fall, can cause considerable leaf loss through direct mortality.

Daughter production and loss rates, as for leaves, were not significantly different between nutritional treatments (Figure 4). As with leaf production rates, daughter production was related to temperature, with an average of one daughter produced every 2 weeks at 30 °C. As with leaf loss, daughter loss was not related to temperature, and averaged

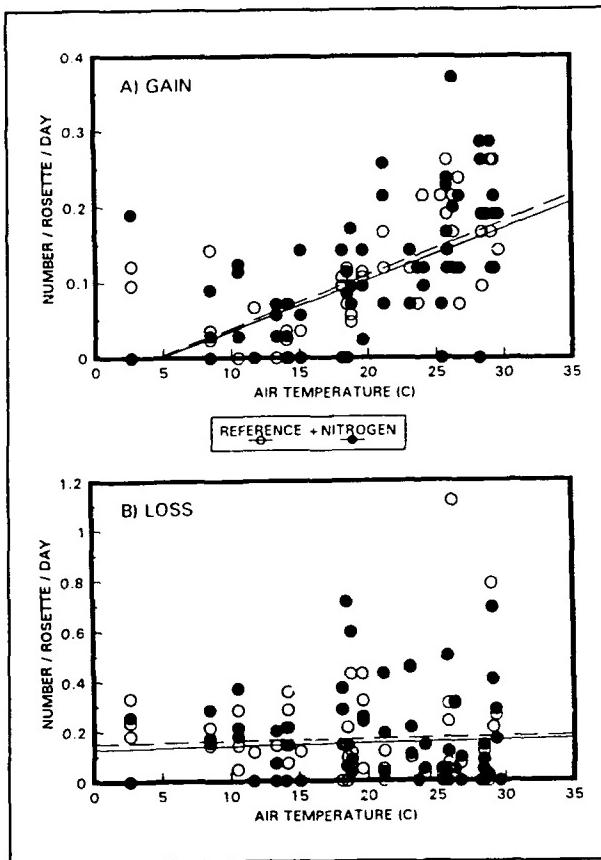


Figure 3. Waterhyacinth leaf gain (A) and loss (B), as measured by leaf tagging, for plants from all cohorts in both reference and nitrogen ponds, expressed as a function of average weekly air temperature (°C)

one daughter plant lost per rosette every 25 days throughout the growing season.

Summary of Results from Current Studies

Temperature was considered the primary environmental factor, controlling rates of photosynthesis, respiration, and leaf and daughter production. However, temperature did not appear to control leaf and daughter loss rates. Nutritional status did not affect the physiological or production processes investigated here, but were important in other parameters, including plant biomass allocation patterns.

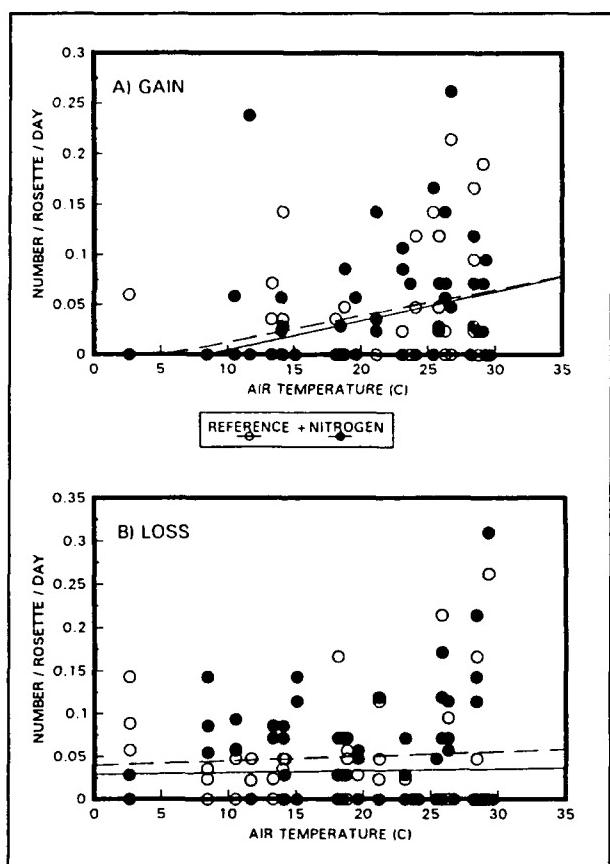


Figure 4. Waterhyacinth daughter gain (A) and loss (B), as measured by daughter tagging, for plants from all cohorts in both reference and nitrogen ponds, expressed as a function of average weekly air temperature ($^{\circ}\text{C}$)

Future Work

Future research on waterhyacinth growth and development will include more detailed investigation into the relationship of light and temperature to leaf photosynthesis and respiration, and the relationship of both pH and nutrient status to whole-plant growth rates.

Eurasian watermilfoil will be the next target plant for additional research efforts, with examinations of respiration rates of different plant parts at different temperatures, and examination of light and temperature effects on shoot photosynthesis. Growth experiments will include detailed examinations of temperature and light effects on the depth limits of plant growth.

Importance of Digital Database Technology to Aquatic Plant Control Simulation Modeling

by

M. Rose Kress¹ and E. May Causey¹

Introduction

The operational use of aquatic plant simulation models is enhanced significantly by the integration of numerical models with digital database technology. Digital database technology provides several unique capabilities to the overall simulation technology thrust area of the Aquatic Plant Control Research Program (APCRP). Some of these capabilities are:

- Organization and management of model input data.
- Organization and access to large volumes of model output data.
- Analysis and visualization of model output data.
- Mapping of model output data over complete study areas.
- Automation of certain aspects of the simulation process.

Geographic information system (GIS) technology is directly applicable to the APCRP simulation modeling task area. To illustrate this, a section of Lake Guntersville, Alabama, was used to develop a GIS database and to develop procedures for using GIS technology with the plant growth models and the HARVEST model. Figure 1 is a navigation chart for the area used to develop a digital database at Guntersville Reservoir.

GIS Data for Plant Growth Models

This section describes how a GIS is used to (1) organize and manage input data for the plant growth models, (2) map the output data from the plant growth models to the appropriate subareas of the study site, and (3) analyze and visualize plant growth model output data. Generally, the plant growth models are structured to make a prediction (or simulation) for one set of conditions (e.g., one site representing one plant type, water depth, water quality, water temperature). For a different set of conditions, the user must supply a new set of input parameters and execute the model again to generate plant growth output data for those conditions.

With the aid of GIS technology, input files for multiple sets of key environmental variables can be generated in an automated fashion and provided/transferred iteratively to the plant growth models. In general, the procedure is to determine all combinations of plant type and water depths present in the study area using GIS technology. This information is transferred to the plant growth models, and model execution is repeated automatically until biomass predictions are made for all existing plant type/water depth combinations in the site or study area. All model output data are stored for later retrieval by the GIS for further analysis of model results or visualization of output data.

¹ US Army Engineer Waterways Experiment Station, Vicksburg, MS.

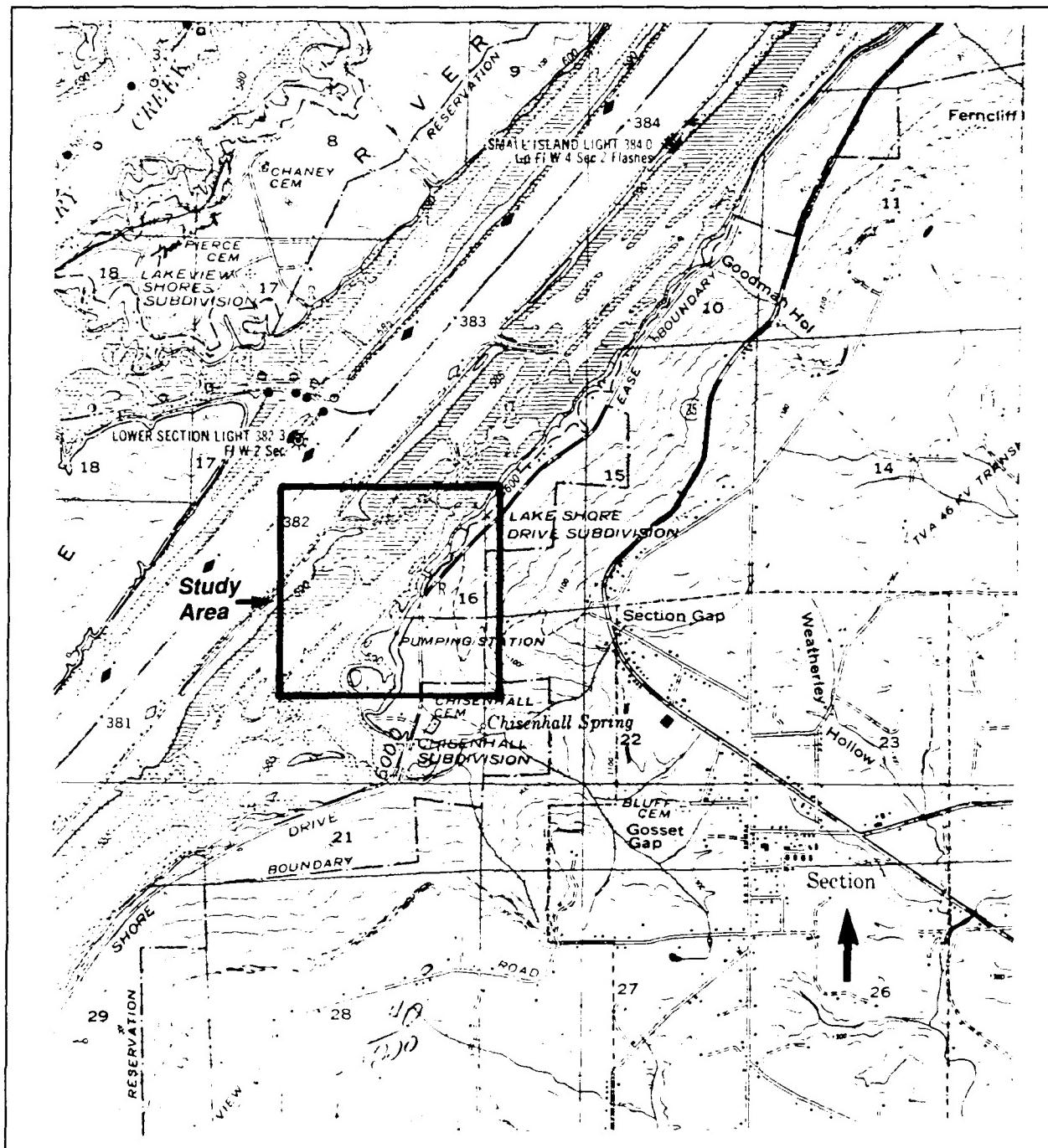


Figure 1. Portion of navigation chart 503 at Lake Guntersville showing location of study site

Database Contents

The relevant data stored in the GIS describing the water body were water depth and plant infestation distribution. The primary source of water depth data was navigation charts. Lake bottom elevations were digitized

from the navigation chart, entered into the digital database, and converted to 5-ft water depth intervals based on an assumed pool elevation of 595 ft mean sea level. Figure 2 is a plot of the resulting digital water depth data. Islands and land areas are also delineated and depicted in the figure.

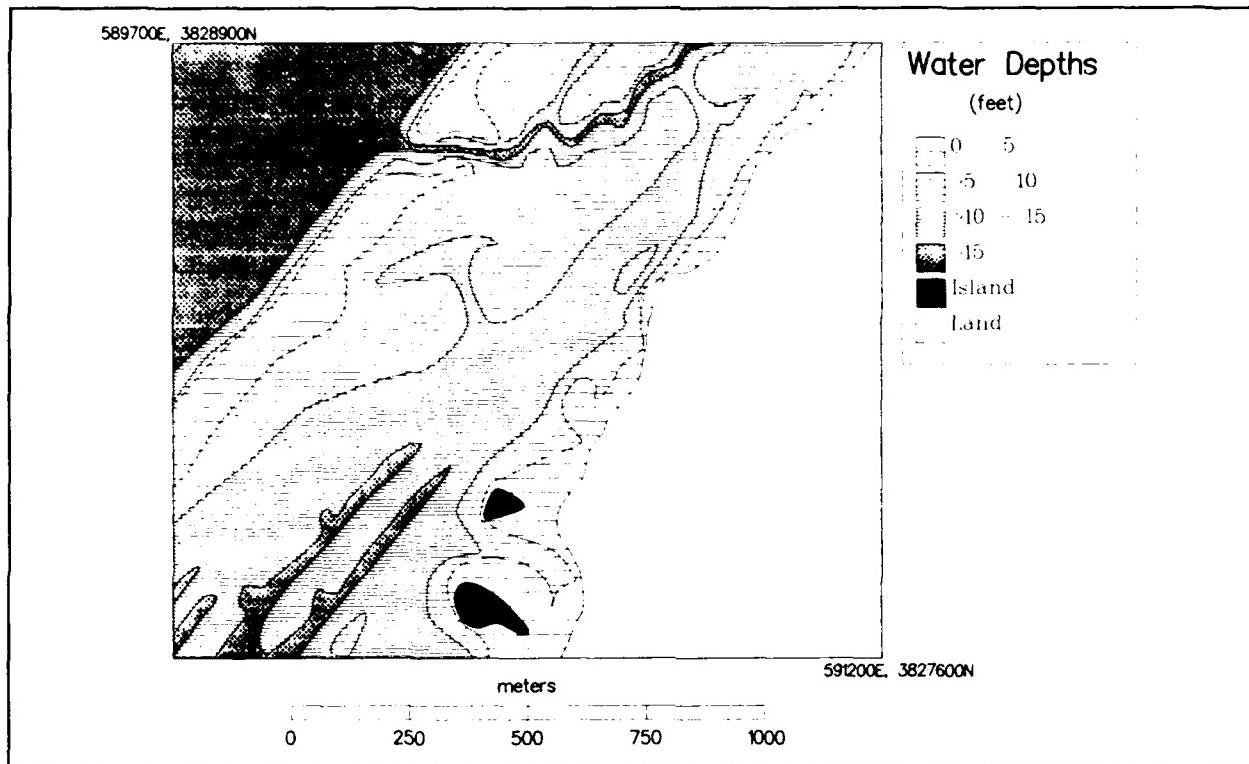


Figure 2. Water depths in 5-ft intervals as determined from navigation chart 503 for the study area

Plant infestation distributions at Lake Guntersville are mapped annually using photogrammetric methods. These maps were digitized into the GIS and coregistered to the water depth data. Currently, predictive growth models are under development for *Hydrilla verticillata* (hydrilla) and *Myriophyllum spicatum* (Eurasian watermilfoil). The plant distribution data in the study area were generalized to show only these two plant types and combinations thereof. Figure 3 is a plot of the resulting plant infestation data.

To identify all combinations of plant type and water depth, the data depicted in Figures 2 and 3 were combined using the overlay function in the GIS. The result of the overlay operation was a new composite data file describing plant type and water depth together. Figure 4 is a plot of the plant type/water depth data. Nine plant/depth classes were identified in the study area, each representing an input condition for the plant growth and HARVEST models.

The appropriate plant growth model (hydrilla or milfoil) was used to predict 2 years (730 days) of daily biomass values for each of these nine unique site conditions. The biomass predictions for mixed plant type conditions were calculated by a weighted average of the corresponding single plant predictions. A total of 6,570 individual biomass predictions (nine classes times 730 days each) were computed. The output data were stored for access by the GIS.

The GIS was then used to map these biomass predictions to all appropriate locations in the study area according to the nine plant/depth classes. Figure 5 is a biomass map generated by the GIS for Julian day 182 (July 1) using output data from the plant growth models. Similar maps were constructed for each of the 730 days of output data. These individual maps were then linked together into a film loop allowing visualization of the predicted seasonal changes in aquatic plant biomass conditions in the study area.

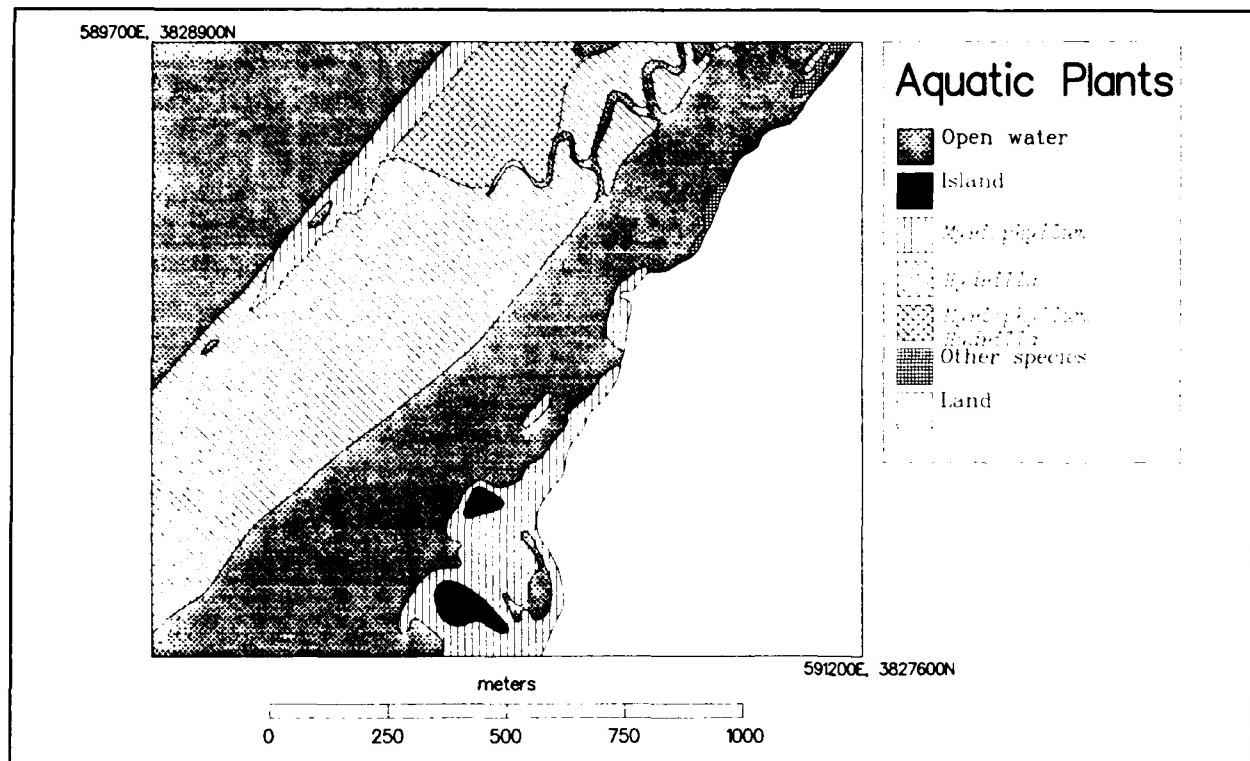


Figure 3. Aquatic plant types in the study area as determined by photointerpretation

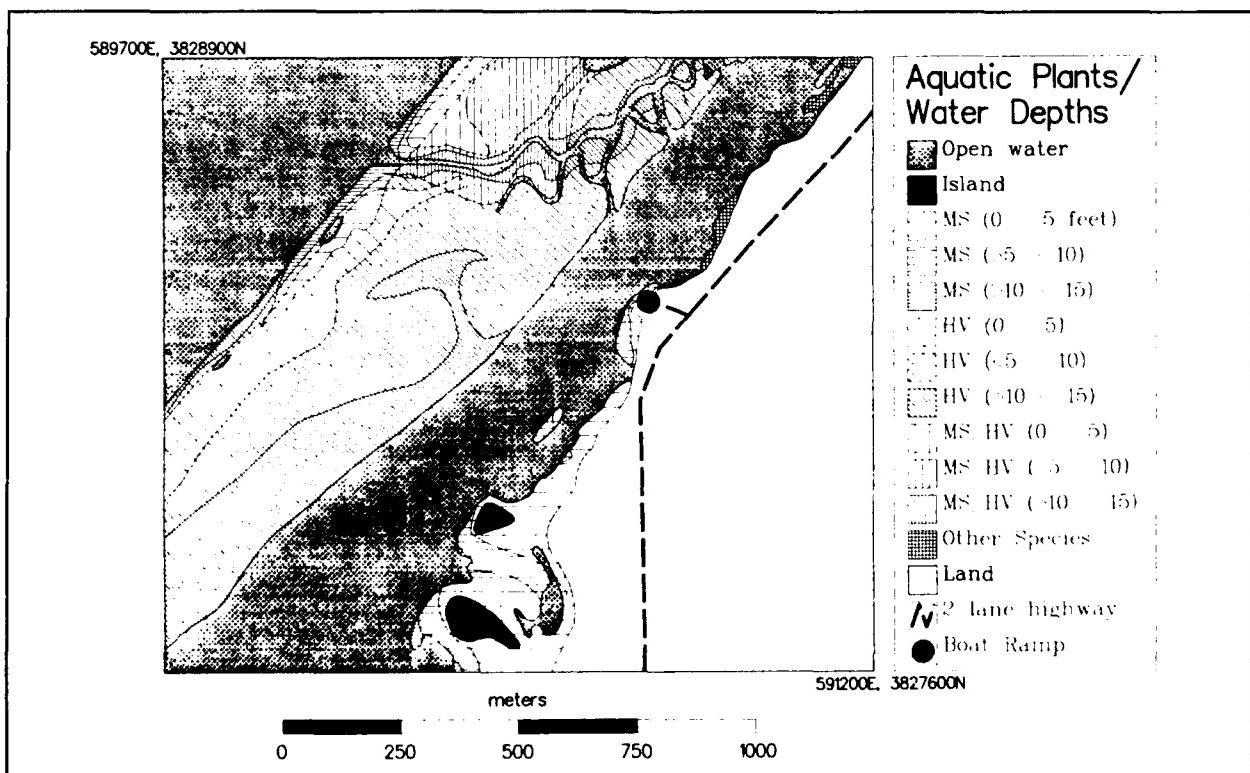


Figure 4. Combined aquatic plant type and water depth classes as determined by GIS analysis
(MS = milfoil; HV = hydrilla)

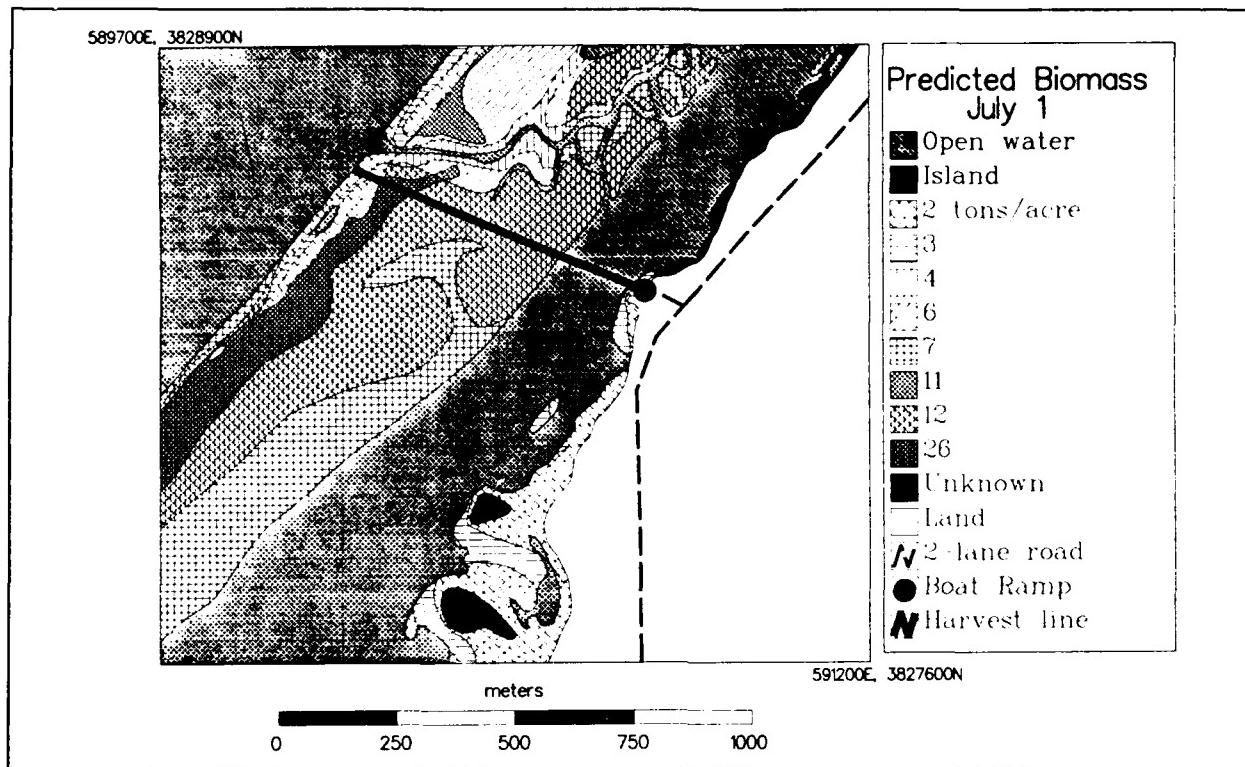


Figure 5. Aquatic plant biomass (tons/acre) as calculated by the plant growth simulation models and presented in map format using the GIS

GIS Data for Use with HARVEST Model

The primary input for the HARVEST simulation model that can be stored and retrieved from the GIS is the aquatic plant biomass of the reservoir area. The previous section discussed how biomass values were predicted by the plant growth models and stored in the GIS. An additional HARVEST model input, length of the harvest site, can also be determined from the GIS.

In Figure 5, a line indicating a user-selected location for a proposed boat lane, running from the boat ramp out to the open channel, is shown. This user-selected boat lane is the location to be modeled by HARVEST, and was selected by the user on the computer screen using a mouse pointing device.

The selected harvest site crosses several areas of differing aquatic plant biomass values. To estimate the total effort and cost to

mechanically harvest the plant material within the selected harvest site, HARVEST requires the length and biomass of each segment. The GIS analysis tools were used to provide this length and biomass information for the HARVEST model.

Table 1 lists the computed length and plant biomass (as predicted previously by the plant growth models) for each segment of the proposed harvest site, as determined by the GIS. The segments as listed in Table 1 begin at the boat ramp and continue out to the open channel, as depicted in Figure 5.

This length/biomass information was used as input to the HARVEST model. If the HARVEST model output indicates the effort and cost are too high, the user can select an alternate location for the boat lane and repeat the simulation.

Table 1
Proposed Boat Lane Description

Segment	Water Depth Class ¹	Length m	Biomass, tons/acre ²
1	MS (>5-10)	3	3
2	Open water	161	0
3	HV (>5-10)	339	12
4	HV (0-5)	74	26
5	MS (0-5)	12	2
6	MS (>5-10)	22	3
7	MS (>10-15)	27	2

¹ MS = milfoil, HV = hydrilla; water depth in feet.
² In tons per acre on 1 July, as calculated by the aquatic plant growth simulation models.

Summary

Digital database technology, especially GIS, can improve the operational usefulness of aquatic plant control simulation models.

Capabilities to automate some aspects of numerical model execution, map model output to other areas, and visualize model output, as discussed in this paper, are significant benefits provided by this technology.

Chemical Control Technology

An Overview of the Chemical Control Technology Area

by
Kurt D. Getsinger¹

Introduction

The primary mission of the Chemical Control Technology Team (CCTT) is to develop technology that will improve the management of nuisance aquatic plants using herbicides and plant growth regulators (PGRs) in an environmentally compatible manner. To accomplish this goal, coordination with the chemical industry (primary developers and manufacturers of herbicides and PGRs) and the US Environmental Protection Agency's (USEPA) Pesticide Registration Branch must be maintained. In addition, interaction with other Federal agencies charged with aquatic plant management—the Tennessee Valley Authority (TVA), US Bureau of Reclamation (USBR), and US Department of Agriculture (USDA)—is necessary to coordinate and focus resources on regional and national problems. Finally, cooperation with state and local aquatic plant management agencies and institutional research facilities is maintained to augment the CCTT's laboratory and field research capabilities. A partial list of research cooperators and contractors is provided in Table 1.

The fiscal year (FY) 1991 direct-allotted funds for chemical control research were apportioned among five work units: Herbicide Concentration/Exposure Time Relationships, Herbicide Delivery Systems, Herbicide Application Techniques for Flowing Water, Field Evaluation of New Herbicide Formulations, and Plant Growth Regulators for Aquatic Plant Management. The work unit Coordination of Control Tactics with Phenological Events of Aquatic Plants is also under the direction of the CCTT.

Summaries of these work units, and of a proposed new start for FY 93 (Species-Selective Use of Aquatic Herbicides and Plant Growth Regulators), are given below. Detailed updates of each work unit are provided in other papers found in the Chemical Control section of this proceedings.

Herbicide Concentration/Exposure Time Relationships (32352)

Research in this area is designed to evaluate USEPA-registered herbicides, as well as experimental use permit (EUP) herbicides for aquatic sites. Target plants, such as Eurasian watermilfoil (milfoil) and hydrilla, are treated with various herbicides at selected doses and contact times under controlled-environment conditions. Results from these studies are used to establish concentration/exposure time relationships for each herbicide and target plant. Evaluations have been completed with the herbicides 2,4-D and triclopyr on milfoil, and endothall on milfoil and hydrilla. Evaluations have been initiated with the herbicides fluridone and bensulfuron methyl on milfoil and hydrilla, and this work will continue in FYs 92 and 93.

Herbicide Delivery Systems (32437)

This work unit explores ways to improve herbicide delivery to submersed plants in high water-exchange environments. One line of research focuses on development of controlled-release (CR) carrier systems, such as polymers, elastomers, gypsum matrices, etc. These CR carriers are being engineered to release herbicides at a slow, predictable rate in

¹ US Army Engineer Waterways Experiment Station, Vicksburg, MS.

Table 1
Partial List of the Chemical Control Technology Team's
Research Cooperators and Contractors

Private Sector		
American Cyanamid	Daychem Labs	Nalco Chemical
Aquatics, Unlimited	DowElanco	Rhone-Poulenc
Asgrow Florida	Du Pont	Southern Research Institute
ASCI Corporation	Gilmore Chemical	Uniroyal
Atochem (Pennwalt)	JLB International Chemical	Valent
Controlled Release Systems	Monsanto	Westvaco
Federal, State, and Local Agencies		
Alabama Game and Fish	South Florida WMD	
Citrus County (FL) Aquatic Plant Management Program	Southwest Florida WMD	
Florida Department of Natural Resources (DNR)	TVA	
Georgia DNR	USBR	
Louisiana Wildlife and Fisheries	USDA - Davis	
Pend Oreille County NWCB	USDA - Fort Lauderdale	
St. Johns Wildlife Management District (WMD)	USEPA	
	Washington, DC, Council of Governments	
	Washington State DOE	
Institutions		
Clemson University	University of California - Davis	
Memphis State University	University of Florida	
Purdue University	University of Mississippi	
Wright State University	University of South Florida	

the vicinity of the target plant. Reliable information on effective herbicide concentration/exposure times is critical for the development of CR formulations. These required dose/contact time relationships are being provided in the aforementioned Herbicide Concentration/Exposure Time work unit.

In FY 91, laboratory studies were conducted at the WES to determine 2,4-D, endothall, fluridone, triclopyr, and bensulfuron methyl release rates from conventional and selected CR formulations. In addition, dye/triclopyr release rates were evaluated (in cooperation with TVA and USBR) using a gypsum CR formulation in the TVA hydraulic flumes at Browns Ferry, Alabama. Future delivery system work will include laboratory, flume, and field evaluations of various CR herbicide carriers.

Herbicide Application Technique Development for Flowing Water (32354)

In this research effort, submersed application techniques are developed and evaluated for their ability to maximize herbicide effi-

cacy against target plants, while minimizing the amount of chemical used and the frequency of treatment. Studies are conducted in large, outdoor hydraulic flumes or in field situations that exhibit high water-exchange characteristics. Recent studies have focused on characterizing water movement, and potential herbicide contact time, in submersed plant stands using flowmeters and tracer dyes. Water movement can dramatically impact the dispersion of herbicides from treated plots, as well as the vertical distribution of herbicide in the water column. Results from this research are used by operational personnel to optimize the type and timing of various submersed application techniques.

Water movement studies and evaluations of submersed application techniques, within submersed plant stands, have been conducted in river, canal, and reservoir systems in the states of Alabama, California, Florida, Georgia, Maryland, Tennessee, Virginia, and Washington. Much of this work has been conducted in cooperation with the University of Florida, as well as various Corps of Engineer Districts and state and local agencies. Research in this area will continue to be

conducted in high water-exchange environments throughout the Nation for the next several years.

Field Evaluation of Selected Herbicides for Aquatic Uses (23404)

In this research area, the most effective application techniques and chemical formulations are evaluated under field conditions. These studies are cooperative efforts among chemical companies, Federal and state agencies, universities, and the WES, with the objective of obtaining efficacy and environmental fate/dissipation data on EUP herbicides and/or new formulations of previously registered herbicides. These data are used to prepare field manuals and reports that provide information on the activity, use, and application techniques of aquatic herbicides. Since these field evaluations can involve changes in registration status, site use, or amendments to residue tolerances, coordination with the USEPA is required.

During FY 91, the WES conducted field studies in Washington and Alabama to test the efficacy and off-target dissipation of the herbicide triclopyr, when used to control milfoil. This work was completed with the cooperation of the TVA, the Seattle District, the Washington State Department of Ecology, and DowElanco. Similar field studies will be initiated in FY92 using the herbicides triclopyr, endothall, and bensulfuron methyl.

Plant Growth Regulators for Aquatic Plant Management (32578)

Plant growth regulators offer the potential for slowing the vertical growth rate of nuisance submersed plants, thereby reducing the negative impacts that "topped-out" plants can impose on a water body. Concurrently, the beneficial qualities provided by underwater vegetation (invertebrate and fish habitat, waterfowl food, oxygen production, nutrient sinks, and sediment stabilization) can be retained.

The WES, in cooperation with industry, the USDA, and Purdue University, is presently exploring the potential for using PGRs to manage submersed vegetation. The compounds bensulfuron methyl and flurprimidol are being evaluated for PGR activity on milfoil and both biotypes of hydrilla. These and other PGRs will be evaluated in controlled-environment and outdoor mesocosm systems during FYs 92 and 93.

Coordination of Control Tactics with Phenological Events of Aquatic Plants (32441)

In this work area, seasonal trends in growth, allocation, and vegetative or sexual propagation and spread of target plants are defined. This information is used to identify weak links in the life cycle of target plants for the application of control tactics, either singly or in concert. Significant contributions have already been made in this effort, particularly in studies of waterhyacinth at both the WES and the Lewisville Aquatic Ecosystem Research Facility (LAERF) in Lewisville, TX. Growth, biomass, and resource allocation studies, leaf-tag studies, and sexual propagation studies were conducted on waterhyacinth in FY 91. In addition, regular measurements of field net photosynthesis and respiration of leaves, stembases, and roots were initiated to provide better data for modeling purposes.

During the next several years, growth, biomass, and resource allocation studies, seasonal photosynthesis and respiration, and vegetative and sexual propagation studies will be conducted on milfoil and hydrilla at the LAERF. In addition to studies at the LAERF, cooperative efforts at other localities will help to establish the geographic variability in phenological phenomena and expand the range of phenological topics examined. Phenology studies will create a better understanding of the basic life cycle and biology of target species, providing insight to the key points at which various management tactics can be implemented.

Species-Selective Use of Aquatic Herbicides and Plant Growth Regulators

The spread of weedy species, such as hydrilla and milfoil, in large water bodies often displaces desirable native plants. While these weedy species can be removed using traditional chemical control tactics, these treatments usually impact native species as well. Furthermore, when aquatic vegetation recovers following chemical treatment, weedy species often prevail. Using herbicides and/or PGRs in a species-selective manner can result in the control of target vegetation, while enhancing the growth of desirable plants. Allowing desirable species to flourish may slow the reinvasion of weedy species and provide improved fish and wildlife habitat. The objective of this proposed new work is to develop and evaluate species-selective aquatic

plant management practices using herbicides and PGRs.

Studies will focus on species-selective responses to applications (both rate and timing) of selected chemicals. Once responses of weedy and various nonweedy species have been identified, desirable, herbicide-resistant plants can be selected for further evaluation. The most promising chemicals will be applied to mixed plant communities established in the recently constructed mesocosm system (Figure 1) at the LAERF. The LAERF mesocosm system will also be used for verification of laboratory-derived herbicide concentration/exposure time relationships, as well as toxicity studies on nontarget organisms. Results from this work will provide guidance for species-selective aquatic plant management practices in the field using chemicals (that is, maximize control of target plants; minimize impacts on nontarget plants).

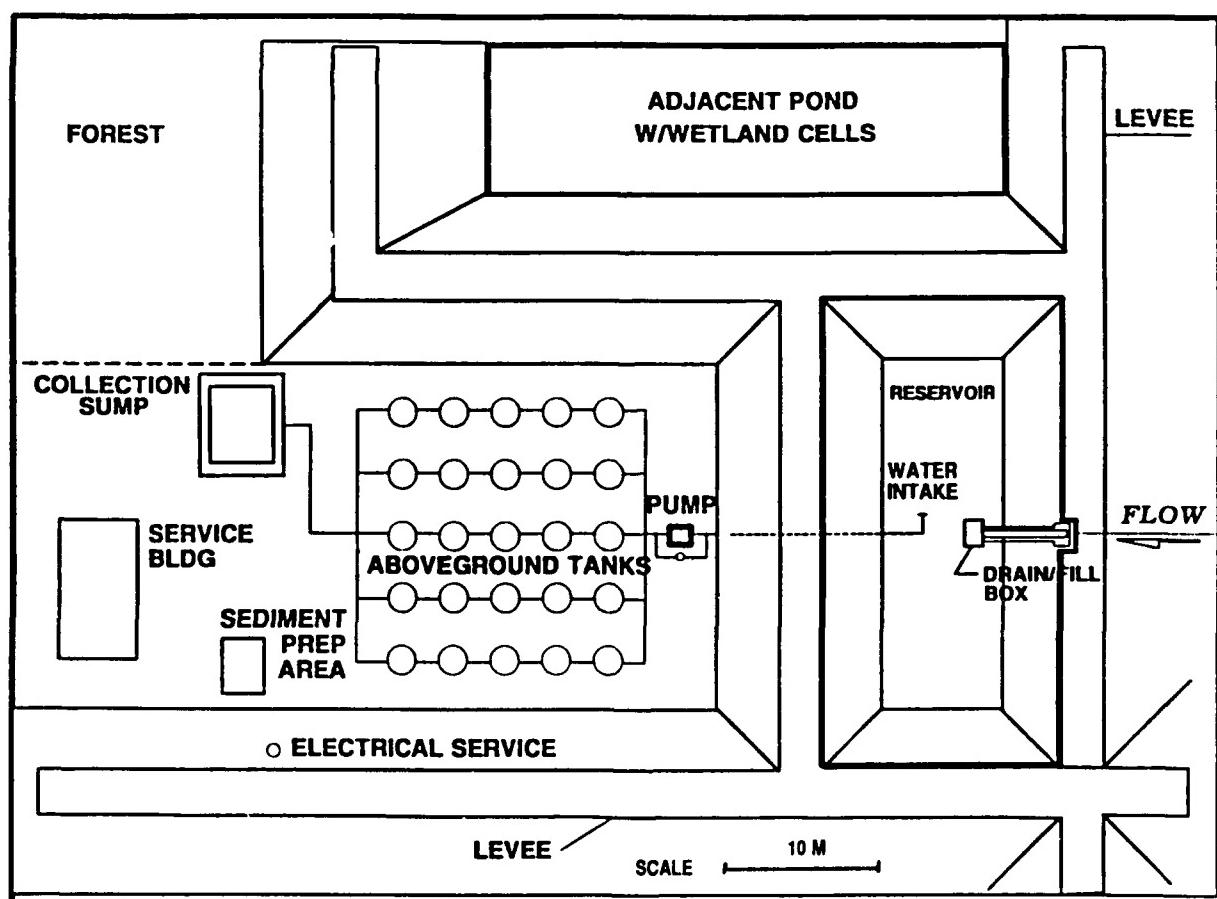


Figure 1. Schematic of chemical control mesocosm system under construction in Lewisville, TX

Herbicide Concentration/Exposure Time Relationships for Eurasian Watermilfoil and Hydrilla

by

Michael D. Netherland¹

Introduction

Following a herbicide application for submersed weed control, physical conditions such as gravity flow, tides, and thermal and wind-induced circulation patterns can rapidly dilute and disperse herbicide residues from the treatment area (Fox et al. 1991, Getsinger et al. 1991). This rapid residue dissipation may result in a lack of plant control due to insufficient herbicide contact time. To assess the effect of rapid residue dissipation on efficacy of submersed applications, laboratory studies of herbicide concentration and exposure time (CET) interactions are being conducted at the US Army Engineer Waterways Experiment Station (Netherland 1991). Results indicate that an increase in the duration of exposure to a given concentration of herbicide is directly related to an increase in plant control.

Recent laboratory studies have included the development of CET relationships for triclopyr versus Eurasian watermilfoil (*Myriophyllum spicatum* L.) and fluridone (Sonar) versus Eurasian watermilfoil and hydrilla (*Hydrilla verticillata* (L.f.) Royle). Triclopyr is currently under an experimental use permit, and data concerning its effectiveness against Eurasian watermilfoil (hereafter called milfoil), particularly in high water-exchange environments, is limited. In contrast, the herbicide fluridone has been registered and successfully used for milfoil and hydrilla control for several years; however, the inconsistency of results in areas of high water exchange remains a problem for applicators using fluridone. The development of CET information will provide guidance for operational

personnel in choosing the herbicide that is best suited for their management objective.

Objective

The objective of this work unit is to identify, in the laboratory, the effective ranges of aquatic herbicide concentrations and exposure times that control milfoil and hydrilla.

Material and Methods

All studies were conducted in controlled-environment systems previously described by Netherland (1990). Temperature was maintained at $24 \pm 3^{\circ}\text{C}$, with a photoperiod of 14L:10D and mean photosynthetically active radiation of $654 \pm 98 \mu\text{E m}^{-2} \text{ sec}^{-1}$. Treatments (herbicide concentration \times exposure time) were replicated three times and randomly assigned to 55-L aquaria. Each aquarium was independently supplied with a continuous flow of water except when herbicide exposures were being conducted.

Four apical tips (10 to 15 cm) were planted in 300-ml beakers containing sediment (amended with nutrients) obtained from Browns's Lake, Vicksburg, MS. A thin layer of silica sand was placed on top of the sediment to prevent resuspension of sediment. Eleven beakers containing plants were then placed in each aquarium.

Plants were allowed to grow approximately 3 weeks prior to herbicide treatment. This pretreatment growth period ensured the development of a healthy shoot and root system. One beaker was removed from each aquarium

¹ US Army Engineer Waterways Experiment Station, Vicksburg, MS.

prior to herbicide treatment to provide an estimate of treated biomass.

Calculated volumes of herbicide stock solutions were added to the aquaria to provide the desired treatment concentrations. Aquaria were drained and refilled three times at the end of the assigned exposure periods. Water samples were taken immediately after treatment (to verify initial concentrations), at the end of the assigned exposure period (to determine possible herbicide dissipation over time), and after the final rinse to verify residue removal.

Plants were allowed to grow 6 to 8 weeks posttreatment, and control was determined by comparing the harvested biomass (separated into shoots and roots) obtained from each treatment. Weekly visual evaluations were used to characterize the initial response and progression of injury symptoms of the treated and untreated plants.

Results and Discussion

Triclopyr is auxin-type systemic broadleaf herbicide with a mode of action and spectrum of weed control similar to that of phenoxy herbicides such as 2,4-D (Weed Science Society of America, WSSA 1990). Milfoil response to triclopyr was characteristic of auxin-like growth regulators. Epinasty occurred rapidly, with apical leaves bending downward and shoots bending and twisting; epidermal rupture began within 36 hr post-treatment. Rapid and serious injury of existing milfoil shoot biomass occurred at most concentrations and exposure times tested. The majority of harvested biomass was the result of shoot regrowth from rootcrown and injured stems. The ability of plants to recover from treatment and produce new healthy biomass decreased as exposure times were increased within a concentration (Figure 1). Treatments of 2.5 mg/L for 2 hr, 1.0 mg/L for 6 hr, and 0.25 and 0.5 mg/L for 12 hr produced initial injury symptoms; however, rapid regrowth indicated the ineffectiveness of these treatments. Root biomass (data not shown) also decreased as exposure times

were increased within a given triclopyr concentration.

Data obtained from harvested biomass were used in conjunction with visual injury assessments to produce a graph that helps predict plant injury based on combinations of triclopyr concentrations and exposure times tested.

Biomass results indicate that triclopyr CET combinations of 0.25 mg/L for 72 hr, 0.5 mg/L for 48 hr, 1.0 mg/L for 36 hr, 1.5 mg/L for 24 hr, and 2.0 and 2.5 mg/L for 18 hr are required to achieve >85 percent reduction of milfoil biomass (Figure 2). As presented in the graph, CET combinations that fall below these levels result in decreasing levels of plant control.

Studies with fluridone differ from triclopyr studies in that exposure times are measured in weeks instead of hours, and concentrations are measured in parts per billion ($\mu\text{g}/\text{L}$) instead of parts per million (mg/L). Hall, Westerdahl, and Stewart (1984) reported that up to 70 days of continuous exposure to fluridone at concentrations >20 $\mu\text{g}/\text{L}$ was required to achieve >85 percent hydrilla control and >95 percent milfoil control (Figure 3). While these studies showed the effects of long-term continuous exposure to low concentrations of fluridone, they did not show the effects of shorter term exposures followed by a regrowth period. Current studies are aimed at filling data gaps by further defining fluridone CET combinations necessary to control milfoil and hydrilla.

Results of short-term static exposures of fluridone on hydrilla indicate that fluridone at 25 and 50 $\mu\text{g}/\text{L}$ for 21 days of exposure was inefficient in significantly reducing hydrilla biomass (Figure 4). New growth of treated plants remained bleached and necrotic while in contact with fluridone; however, upon fluridone removal, plants began to regrow from rootcrown and lower stems. Results indicate that at fluridone concentrations <50 $\mu\text{g}/\text{L}$, exposure times of >21 days will be required to control hydrilla.

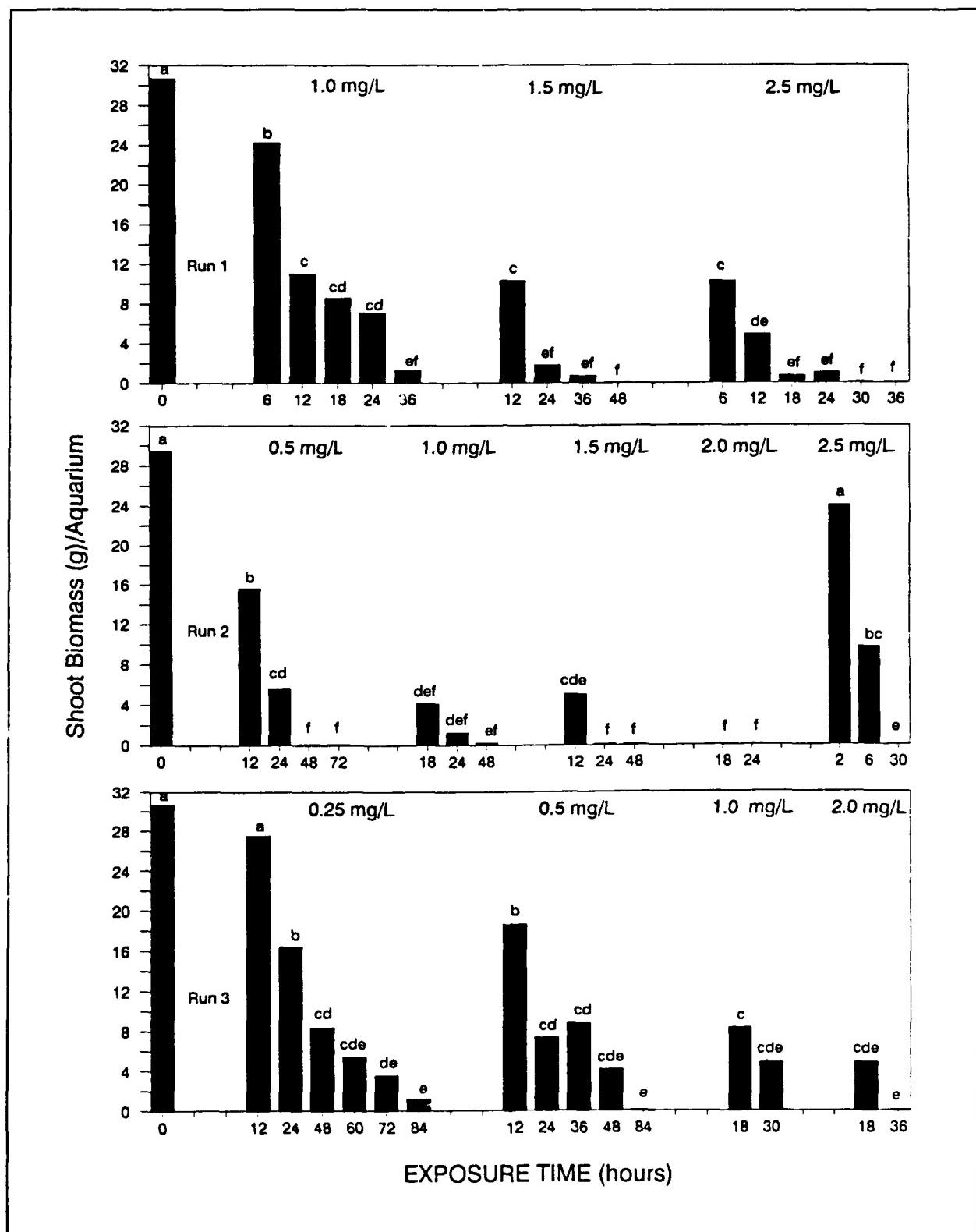


Figure 1. Eurasian watermilfoil shoot biomass at 6 weeks posttreatment with triclopyr.
 Data are the means of three replicates; different letters within a treatment run indicate significant differences at the 5-percent level

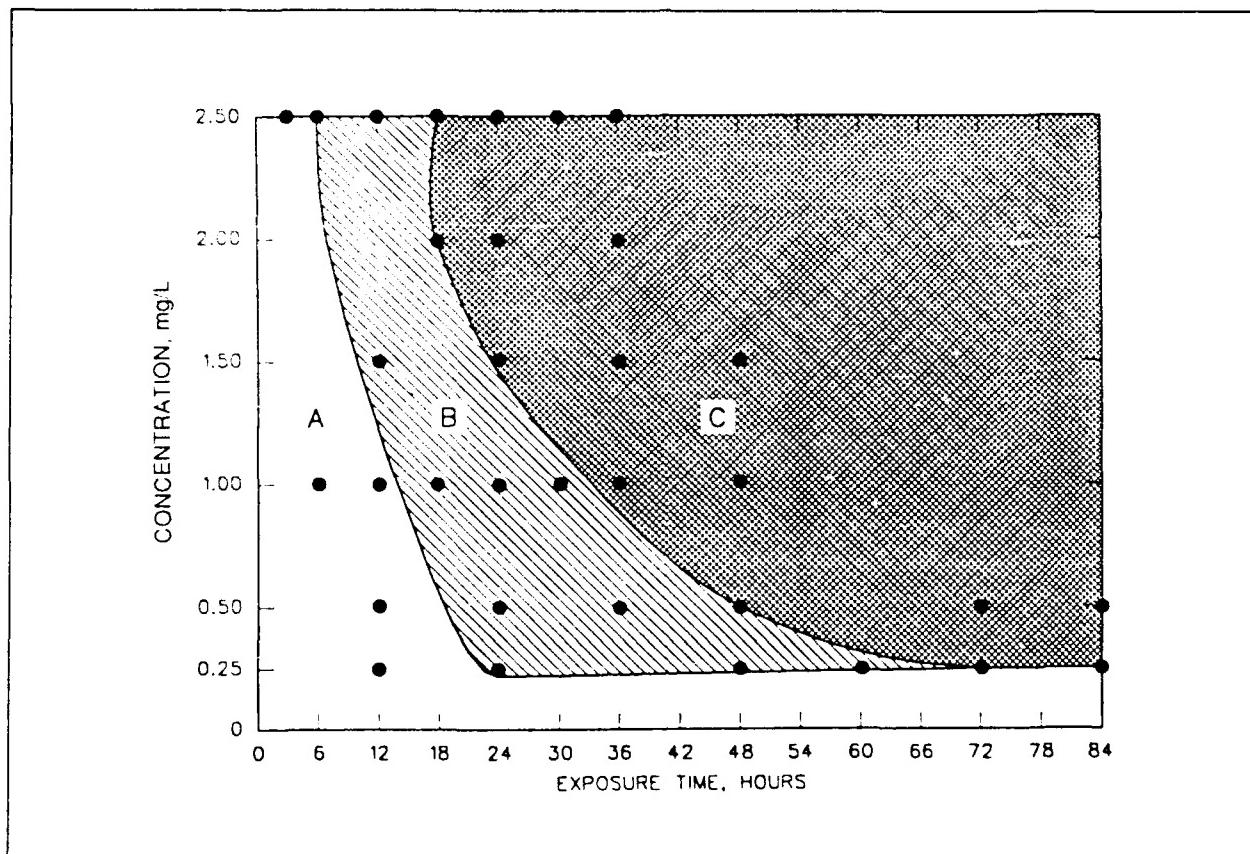


Figure 2. Summary figure of triclopyr concentration/exposure time relationships for control of Eurasian watermilfoil. Zone C represents CET combinations that provided >85 percent milfoil control; Zone B represents combinations that gave between 70 and 85 percent control; and Zone A represents combinations that gave <70 percent milfoil control

A fluridone/milfoil study was designed using information from the fluridone/hydrilla study. Exposure times were increased to range from 21 to 70 days, while fluridone concentrations ranged from 5 to 24 µg/L. Results indicate that concentrations of 12 and 24 µg/L at 21 days of exposure gave approximately 50 percent milfoil control, while exposure times of 28 to 42 days provided >80 percent milfoil control (Figure 4). Treatments of 12 µg/L at 35 days and 24 µg/L at 42 days were the only treatments that prevented healthy regrowth from rootcrown following removal of fluridone. Results indicate that increased exposure time is the key to improving milfoil control with fluridone.

The direct application of laboratory results to the field should be viewed with some degree of caution. Exposure times in the field

are dynamic in that the plant is exposed to a dissipating concentration of herbicide over time. In addition, differences in recuperative capacity (particularly from rootcrown) and differences in sensitivity between mature field plants and plants grown from cuttings require caution. While difficulty remains in precisely predicting field efficacy from laboratory results, the relationship of increased herbicide concentrations and exposure times to increased plant control has been established.

Information obtained from these CET studies is currently being coupled with other work units within the Chemical Control Technology area. The use of dye studies to determine potential herbicide dissipation represents a new, relatively inexpensive method to predict potential contact time in a variety of aquatic systems (Fox, Haller, and Getsinger

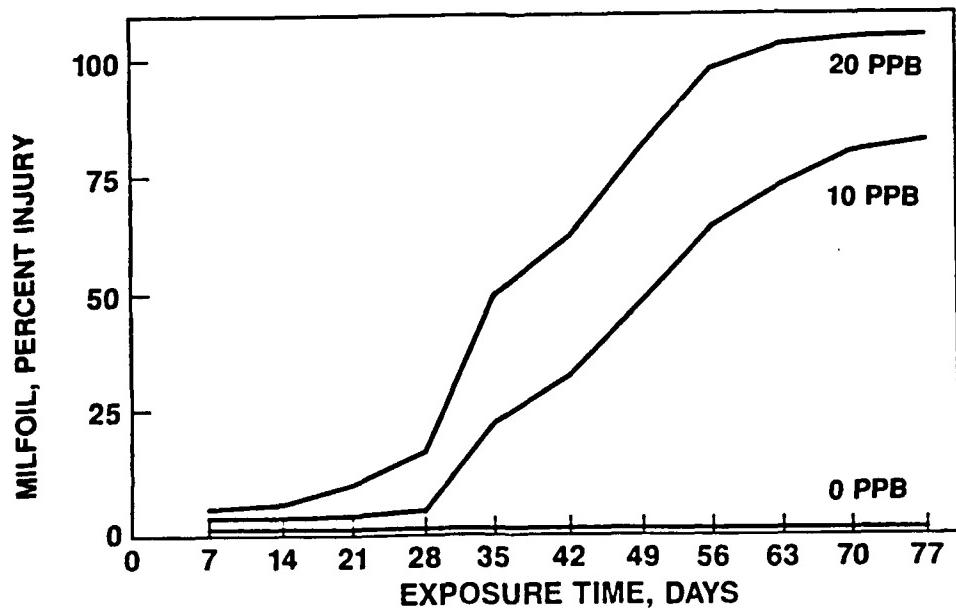
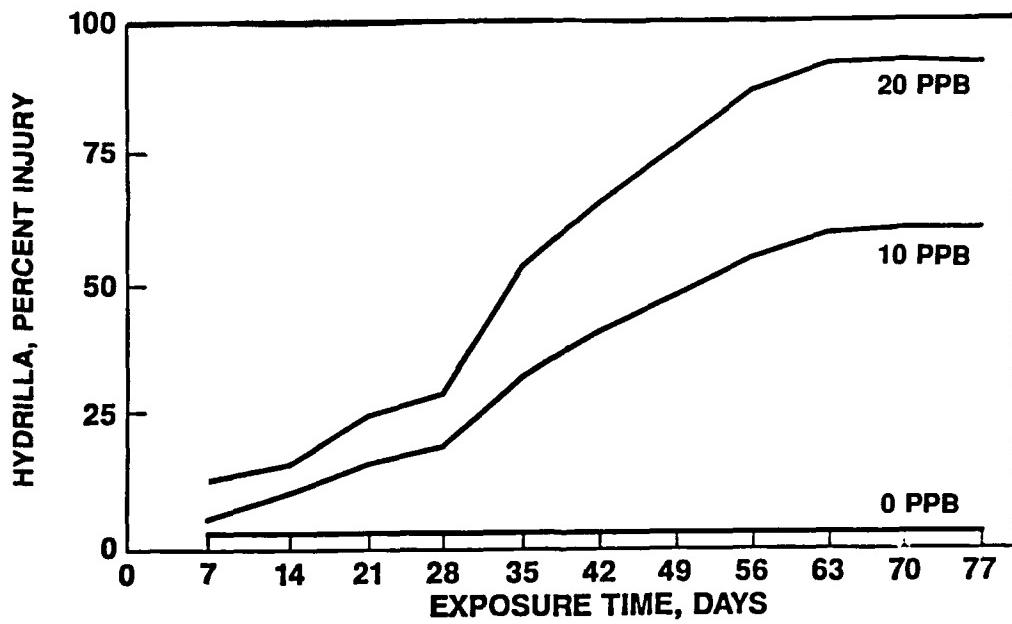


Figure 3. Estimated hydrilla and milfoil percent injury following continuous exposure to 10 and 20 ppb fluridone

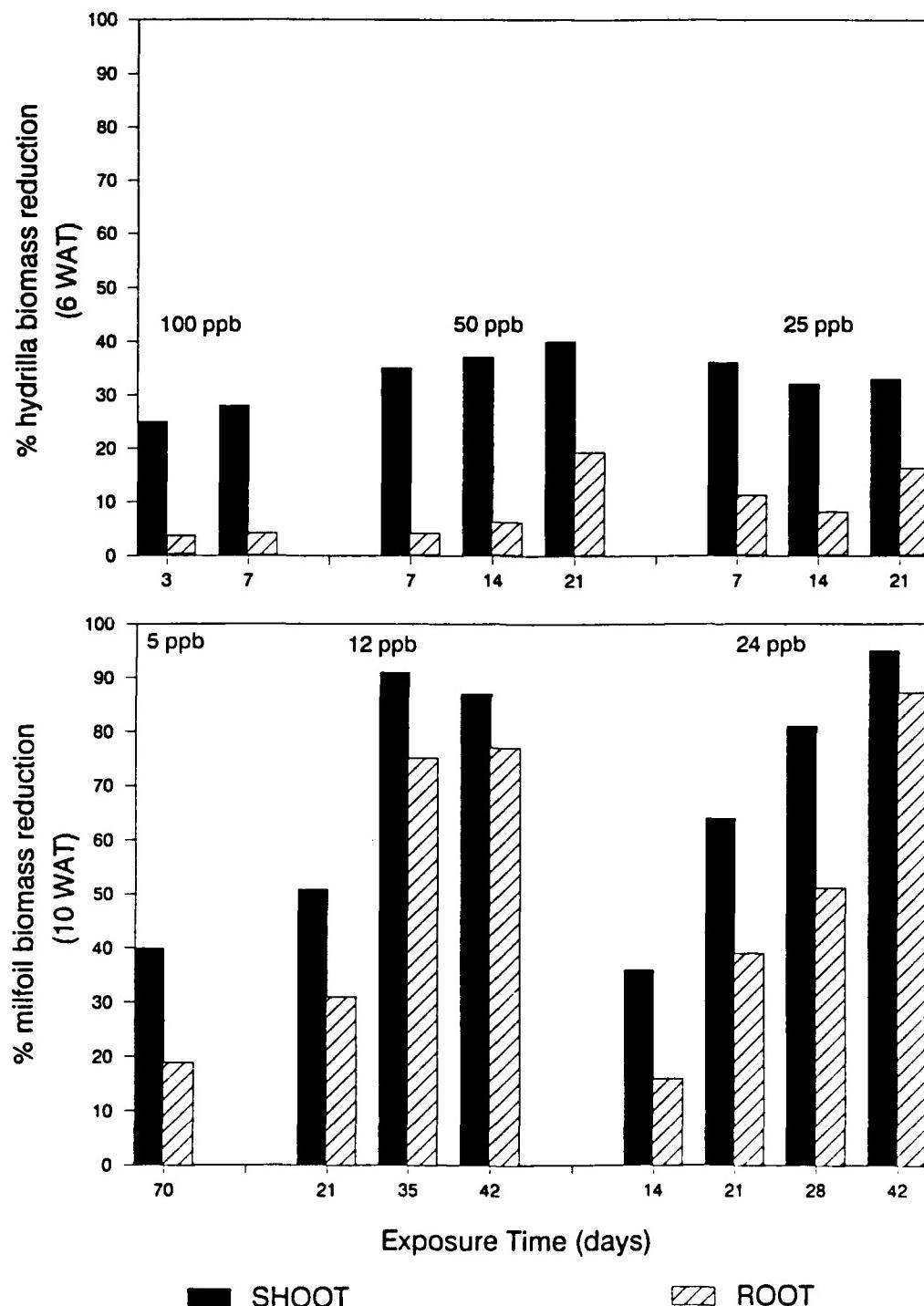


Figure 4. *Hydrilla and milfoil biomass reductions following static exposures to various concentrations of fluridone (WAT = weeks after treatment)*

1990; Fox et al. 1991; Getsinger et al. 1991). Furthermore, to validate laboratory-scale studies, large outdoor CET flume studies are being conducted in cooperation with the Tennessee Valley Authority.

Future work will include further studies with fluridone on hydrilla and milfoil and initiation of studies with bensulfuron methyl on milfoil.

Acknowledgments

The author would like to thank Brian York, Kim Deavers, Sheron Burt, and Glenn Turner for laboratory assistance. The cooperation of DowElanco, Inc., for providing herbicide formulations and residue analyses is also appreciated.

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Herbicide Concentration/Exposure Time Flume Studies

by

E. Glenn Turner,¹, Michael D. Netherland,¹ and Earl R. Burns²

Introduction

Concentration/exposure time (CET) relationships for triclopyr and Eurasian watermilfoil have recently been developed in laboratories at the Waterways Experiment Station (Netherland and Getsinger 1992).³ These studies clearly demonstrate that triclopyr is capable of providing excellent control of Eurasian watermilfoil given the appropriate conditions. As part of a comprehensive effort to provide guidance for the most effective use of triclopyr in the field, these CET relationships were further evaluated in a scale-up study utilizing outdoor hydraulic channels (flumes).

The flume system used in this study provides an experimental situation in which environmental conditions are more characteristic of natural systems than conditions in most indoor laboratory facilities, yet control of certain experimental variables is still maintained. Thus, flume studies help bridge the gap between bench-scale laboratory experiments and large-scale field studies. The work described herein will further the development

of CET relationships and improve accuracy in the application of those relationships to field operations.

Materials and Methods

This study was conducted in eight flow-through, concrete flumes located at the Tennessee Valley Authority Aquatic Research Laboratory (ARL) at Browns Ferry, AL. Each flume measures 112 m in length and 4.3 m in width, and is lined with a 49-cm-thick layer of reservoir sediment (Figure 1). Water is drawn from Wheeler Reservoir, and depths from 0.6 to 1.2 m can be attained by varying the number of weir boards at the outlet end of each flume. At maximum water depth and flow rate, complete water exchange requires 16 to 20 hr.

Stands of Eurasian watermilfoil (hereafter referred to as milfoil) (4.3×12 m) were established in each flume beginning approximately 40 m from the inlet end (Figure 2). This was accomplished by first draining each flume, then planting four to six freshly harvested

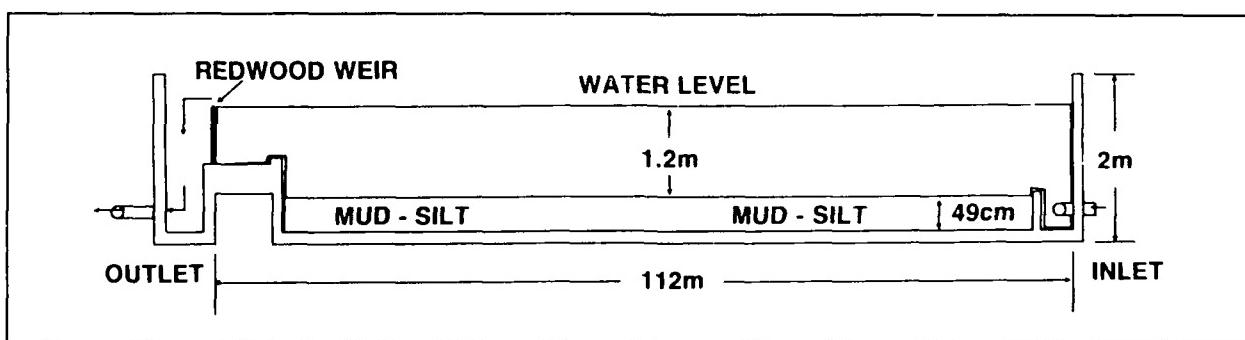


Figure 1. Diagram of ARL flume

¹ US Army Engineer Waterways Experiment Station, Vicksburg, MS.

² Tennessee Valley Authority, Muscle Shoals, AL.

³ M. D. Netherland and K. D. Getsinger. 1992. Efficacy of triclopyr on Eurasian watermilfoil: Concentration and exposure time effects. *Journal of Aquatic Plant Management* 30:1-8.

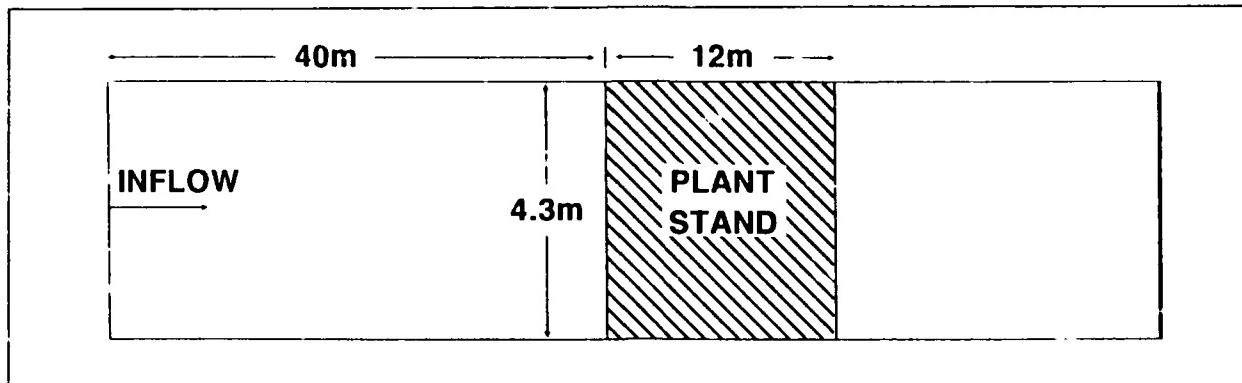


Figure 2. Top view of flume showing location and dimensions of milfoil stand

apical tips, 15 to 20 cm in length, on 0.5-m centers and allowing the plants to grow in flowing water for 5 weeks. Milfoil tips were collected from a heavily infested pond located on the Browns Ferry Nuclear Power Plant facility, adjacent to the ARL. Planting was conducted on June 10 and 11, 1991. Final water depths in the eight channels ranged from 0.92 to 1.06 m. Mean pH and temperature values ($n = 8$) of the inflow water at the time of planting were 7.4 ± 0.2 and 26.9 ± 0.5 °C, respectively.

One day prior to herbicide treatment, water flow to all flumes was terminated, and plywood partitions were installed in seven of the eight flumes (Figure 3). The partitions divided each flume into an upper section (38 m in length) and a lower section (76 m in

length). Milfoil stands were located in the lower section of the seven channels. At this time most plants were at or near the water surface; however, extensive surface mats had not yet developed.

Herbicide applications were conducted on July 17, 1991. Seven triclopyr treatments, each consisting of a unique concentration/exposure time combination, were used (Table 1). These CETs were based on results of previous studies conducted at the WES (Netherland and Getsinger 1992). The remaining untreated flume served as a reference. In addition, the fluorescent dye Rhodamine WT was applied to each flume, concurrent with the herbicide application, at a rate calculated to achieve a final concentration of 10 µg/L.

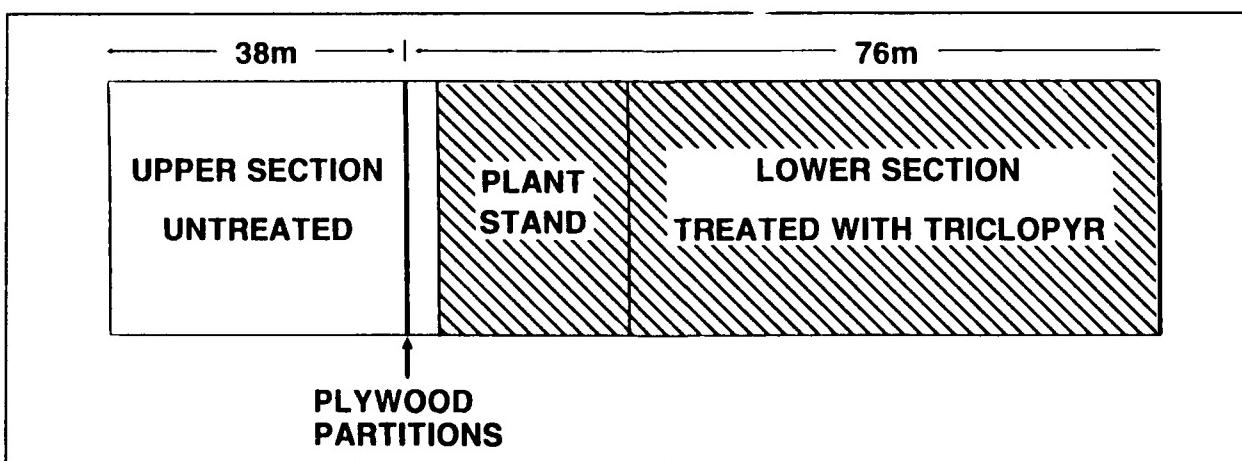


Figure 3. Top view of flume showing location of plywood partitions and dimensions of treated and untreated (hatched) sections

Table 1
Triclopyr Concentrations
and Exposure Times

Treatment	Concentration ¹	Exposure Time hr
Ref	0.0	—
1	0.5	18
2	0.5	36
3	1.0	18
4	1.0	36
5	2.0	18
6	2.0	36
7	2.0	168

¹ Values expressed as milligrams per liter active ingredient.

Applications were accomplished using a submersed injection technique in which the herbicide/dye mix was pumped through eight alternating short and long weighted hoses attached to a 4.3-m boom. Short hoses extended 20 cm below the water surface, and long hoses extended 40 cm below the water surface. Applications were made to the entire volume of the single, nonpartitioned flume, but only to the lower sections of the seven partitioned flumes.

At the end of each 18- and 36-hr exposure period, water from these channels was discharged by removing the partitions and weir boards followed by resumption of water flow. The untreated water in the upper sections of the flumes acted as dilution water, ensuring that triclopyr concentrations within the plant stands were rapidly reduced following the exposure period. After flushing of the flume had been completed, weir boards were replaced and initial water depths re-established. In the 168-hr exposure flume, which was not partitioned, flushing of the herbicide was accomplished only by resuming water flow. This resulted in a slower removal of triclopyr from the plant stand; however, with a 168-hr exposure period, rapid removal was not deemed critical.

During the exposure periods, water samples and dye measurements were taken from two

stations within each plant stand at a depth of 50 cm. During discharge of treated water, water samples and dye measurements were taken from three stations within the plant stands at a depth of 25 cm. Generally, these data were collected concurrently at 1 to 2 hr posttreatment, midtreatment, immediately prior to discharge, and at 2, 10, 20, and 36 hr following discharge. However, as the result of inclement weather, dye concentrations were not measured in some cases. In the 168-hr exposure treatment, postdischarge samples were collected only at 24 and 48 hr. All water samples were frozen for later residue analysis. Dye concentrations and water temperatures were monitored using a Turner Designs model 10-005 field fluorometer equipped with a high-volume continuous-flow cuvette system and a temperature probe. Dye amounts were corrected for temperature variation according to Smart and Laidlaw.¹

Four to six 0.5-m² biomass samples were collected from each flume 1 day prior to herbicide/dye applications and at 7 weeks post-treatment. Plants were washed to remove debris, divided into root and shoot portions, and oven-dried (95 °C) to a constant weight. Triclopyr efficacy was evaluated by comparing pretreatment and posttreatment shoot biomass within each flume, and by comparing posttreatment biomass of each treated flume to that of the reference flume.

Results and Discussion

Herbicide residues

After allowing time for uniform dispersion of the herbicide within the flumes, triclopyr residue data indicated that actual concentrations were within a mean of ± 16 percent of target rates. The only outstanding loss of residue during the treatment periods occurred in the 168-hr exposure flume where the triclopyr concentration was reduced to approximately 30 percent of the target rate by the end of the exposure period. This was presumably the

¹ P. L. Smart and I. M. S. Laidlaw. 1977. An evaluation of some fluorescent dyes for water tracing. *Water Resources Research* 13(1):15-33.

result of a combination of photo/microbial degradation and plant uptake.

In all 18- and 36-hr exposure flumes, herbicide residues were reduced by 97 percent or more within 2 hr following termination of each exposure period. In the 168-hr exposure flume, triclopyr was reduced by 99 percent within 24 hr following the exposure period (the minimum interval at which water samples were collected).

Herbicide efficacy

An initial complete knockdown of milfoil occurred in all flumes treated with triclopyr within 8 to 10 days following herbicide applications. At 7 weeks posttreatment, aboveground regrowth in all treated flumes was limited to 10 percent or less of the pretreatment levels (Figure 4). At a concentration of 0.5 mg/L triclopyr, little difference in efficacy was noted between the two exposure periods; milfoil was reduced from 52.6 ± 6.6 to 5.3 ± 1.2 g dry weight (DW)/m² at an exposure time of 18 hr and from 40.3 ± 6.1 to 3.3

± 1.2 g DW/m² at an exposure time of 36 hr. The posttreatment plant material collected from these treatments consisted of healthy, green shoots that were 15 to 40 cm in length.

Posttreatment biomass in all other treatments was zero with one exception. In the 2.0-mg/L, 18-hr exposure flume, three shoots were found still attached to the roots. This comprised the 0.4 mg DW/m² shown in Figure 4. However, these shoots were completely brown and were not considered viable.

Although there were some differences, these results compare very favorably to those of the WES CET laboratory studies (Netherlands and Getsinger 1992). Table 2 compares the degree of milfoil control predicted from the WES laboratory experiments (conducted in controlled-environment chambers) to actual control levels obtained in the ARL flumes. In all flume treatments, milfoil control levels matched or exceeded those predicted from the laboratory experiments. For all of the 2.0-mg/L treatments and the 1.0-mg/L treatment at 36 hr, predicted control was 85 to

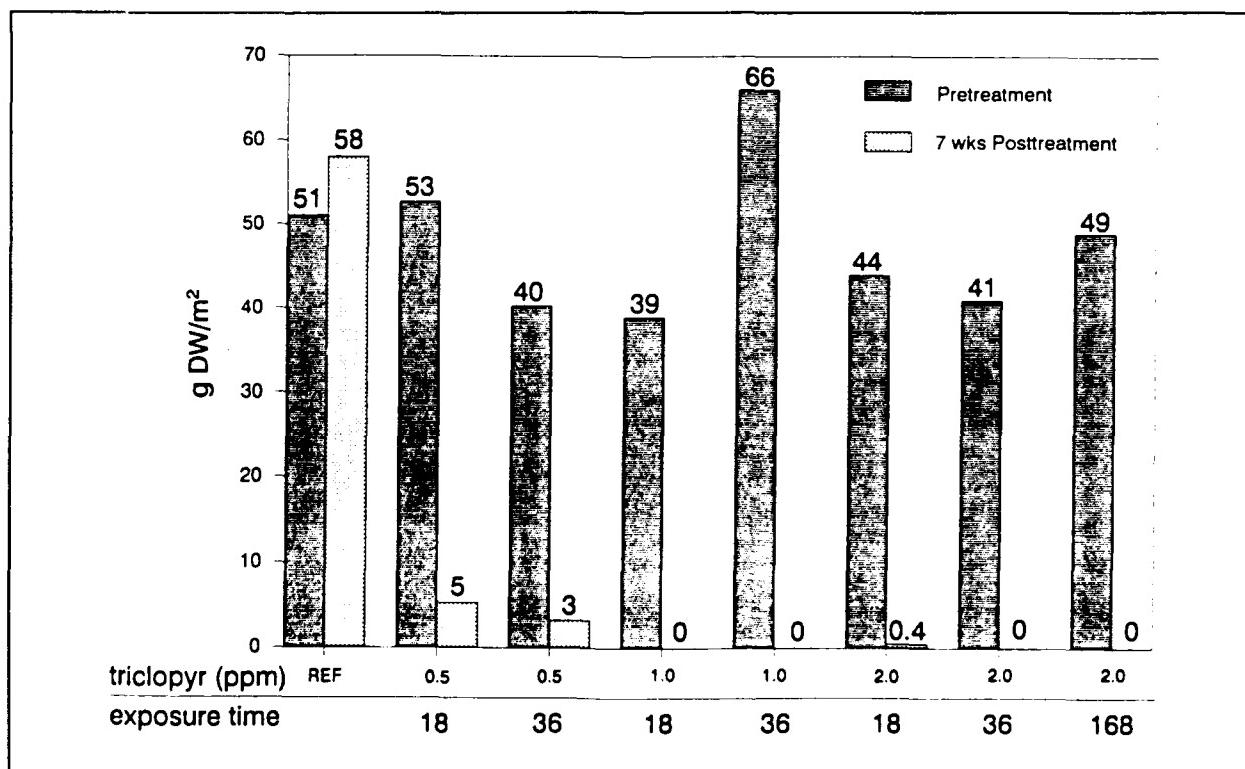


Figure 4. Pretreatment and 7-weeks posttreatment milfoil biomass values

Table 2
Comparison of Predicted and Actual Milfoil Control Levels

Concentration ¹	Exposure Time, hr	Percent Control	
		Predicted	Actual
2.0	168	100	100
2.0	36	85-100	100
2.0	18	85-100	100
1.0	36	85-100	100
1.0	18	70-85	100
0.5	36	70-85	90
0.5	18	<70	90

¹ Values expressed as milligrams per liter active ingredient.

100 percent. Actual control for all four treatments was 100 percent. Predicted control was 70 to 85 percent for the 1.0 mg/L, 18-hr exposure treatment, and actual control was 100 percent. For both of the 0.5-mg/L treatments actual control was 90 percent, whereas predicted control was lower—70 to 85 percent for the 36-hr exposure period and <70 percent for the 18-hr exposure period.

Results from this study showed that triclopyr concentrations and exposure times predicted from controlled-environment studies were effective in controlling milfoil in outdoor flumes where conditions were more similar to natural systems. Differences between predicted and actual control from lower concentrations were not unexpected,

since herbicide efficacy and aquatic plant regrowth are influenced by environmental conditions such as temperature, light, and plant growth stage. This emphasizes that while experiments conducted in controlled environments are necessary in establishing concentration/exposure time relationships, field testing under a range of environmental conditions is also necessary to determine rates and use guidelines for most efficacious control.

Future Research

Plans for future flume studies at the ARL include evaluations of controlled-release herbicide delivery systems, further verification of laboratory CET studies, and evaluations of herbicide selectivity within mixed-plant communities.

Acknowledgments

The authors wish to thank DowElanco for triclopyr residue analysis and the following people for technical assistance: Tommy Woods, Carl Wilmer, Rick Johnson, and Dan Harraway, Tennessee Valley Authority; Mike Crouch, Lewisville Aquatic Ecosystem Research Facility, Lewisville, TX; and Anne Stewart, ASci Corporation, Vicksburg, MS.

Aquatic Herbicide Delivery Systems

by

Michael D. Netherland¹

Introduction

The key to successful control of submersed plants with chemicals in high water-exchange environments is maintaining an adequate herbicide exposure period. Ross and Lembi (1985) stated that the major prerequisites for effective herbicide use are that a sufficient amount of herbicide come in contact with the plant surface, remain at the plant surface long enough to penetrate or be absorbed into the plant, and reach a living, cellular site where it can disrupt a vital process or structure. Information obtained from the Herbicide Application Technique work unit (Fox, Haller, and Getsinger 1990; Fox et al. 1991; Getsinger, Haller, and Fox 1990; Getsinger et al. 1991) and the Herbicide Concentration/Exposure Time work unit (Van and Conant 1988; Green and Westerdahl 1990; Netherland, Green, and Getsinger 1991; Netherland and Getsinger 1992) suggests that a lack of chemical contact time may be responsible for the failure of many herbicide treatments.

It is clear that a need exists to develop herbicide formulations and/or delivery techniques that will extend chemical contact time to improve the control of submersed vegetation in high water-exchange environments.

One approach for extending contact time is to develop a controlled-release (CR) carrier or matrix. A CR matrix or formulation is defined as an active ingredient of a pesticide (herbicide, insecticide, plant growth regulator) combined with an inert carrier (polymer, lignin, clay, etc.). Riggles and Penner (1991) state that the potential benefits of controlled release include enhanced weed control, reduced cost, less environmental impact, and greater safety in handling.

Developing a CR formulation requires data that define the unique concentration/exposure time (CET) requirements of a herbicide against the target species. Data obtained from CET studies indicate that as exposure time is increased, lower concentrations of herbicide can be used to achieve plant control (Figure 1).

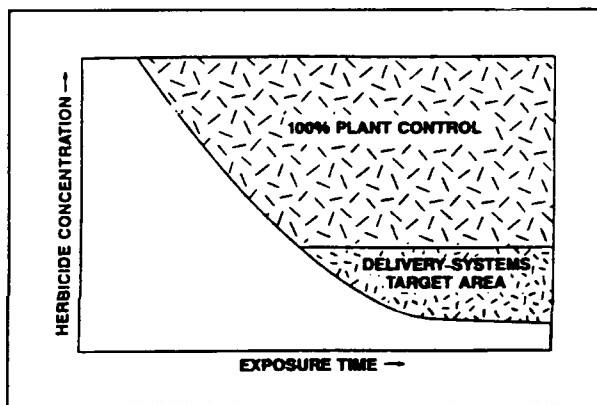


Figure 1. Proposed target area for slow-release delivery systems. (Note the requirement for lower herbicide concentrations and increased exposure time)

Information concerning the internal herbicide tissue burden should also aid in the development of CR matrices. Several researchers have shown that while tissue concentrations of herbicides are much higher than concentrations in the ambient water, only a small proportion of the herbicide in the water is actually taken up by the plant (Table 1). These results indicate that, by extending the exposure period, lower rates of herbicide could be used with improved efficacy.

It should be noted, however, that external concentrations must be sufficient to obtain the critical level of herbicide that must be

¹ US Army Engineer Waterways Experiment Station, Vicksburg, MS.

Table 1
Internal Tissue Levels of Herbicides Following Treatment

Investigators	Herbicide	Target Species	Herbicide in Plant Tissue (% of total applied)
Van and Conant (1988)	Endothall	Hydrilla	~7
Haller and Sutton (1973)	Endothall	Hydrilla	~6
Van and Steward (1986)	Fluridone	Hydrilla	~3
Reinert et al. (1985)	Endothall	Milfoil	~3
Marquis, Comes, and Yang (1981)	Fluridone	<i>Potamogeton</i> spp.	~0.6

reached within the plant tissue to achieve a lethal dosage. The concentration of the herbicide at a vital location in the plant at any one given time may determine the herbicide effectiveness, whereas the same quantity of herbicide over an extended period of time may have little or no effect (Klingman and Ashton 1982).

Many CR herbicide matrices have been evaluated during the last 20 years (Steward and Nelson 1972; Van and Steward 1982, 1983, 1986; Connick et al. 1984; Harris 1984; Dunn et al. 1988) with mixed results. Current work focuses on new matrices used in aquatic insect control. Gypsum- and protein-based products have been developed to release insecticides for a period of up to 60 days. The proven slow-release characteristics and environmental compatibility of these compounds make them excellent candidates for CR herbicide matrices.

Increased environmental concern by the public has stimulated a demand for reduced loading of pesticides into the environment. Developing new methods to deliver herbicides in the most efficient manner to target plants offers the potential of using less active ingredient to achieve greater efficacy. This approach will ultimately translate into superior herbicide efficacy at lower treatment rates, resulting in enhanced environmental compatibility.

Objectives

The objectives of this work unit are to evaluate and develop delivery systems that

will maximize herbicide contact time against submersed macrophytes within a treatment area.

Research Approach

Herbicide delivery systems will be evaluated over the next several years, with initial efforts focusing on CR matrices and the effects of repeat applications in flowing water systems. The following approach will be used to initially evaluate CR matrices:

- Environmentally compatible CR matrices will be identified and selected for evaluation, with priority given to matrices proven for slow release of pesticides in an aquatic environment.
- Matrices will be formulated with a variety of aquatic herbicides to test for compatibility between the matrix and the active ingredient of the herbicide.
- Successfully formulated herbicide matrices will be evaluated in the laboratory for potential controlled-release characteristics and for comparison with release rates of registered conventional slow-release formulations.

Materials and Methods

Gypsum- and protein-based matrices (Controlled Release Systems Research, Inc.) were formulated as 2-percent granules with the herbicides 2,4-D, triclopyr, fluridone, and benzulfuron methyl. Conventional formulations

tested included Aquakleen (19-percent 2,4-D granular), Sonar SRP (5-percent fluridone granule), Aquathol K (7-percent endothall granule), and a new 27-percent endothall granule. Measured quantities of formulations were placed in 55-L aquaria with a water temperature of $22 \pm 2^{\circ}\text{C}$. At the end of each 24-hr period, the aquaria were drained twice and refilled; water samples were taken at 2, 12, and 24 hr for 7 days.

Results and Discussion

Release rate results for fluridone, 2,4-D, and triclopyr are summarized in Tables 2 and 3 (bensulfuron methyl and endothall samples are currently being analyzed). These preliminary data suggest that all matrices were compatible with the herbicides tested and exhibited some controlled-release properties.

The gypsum and protein matrices formulated with fluridone released fluridone at a fairly steady rate during the study, although concentrations never exceeded 10 ppb/day. The Sonar SRP pellet released fluridone at a constant rate for 4 days and maintained concentrations near 15 ppb/day. At the end of

7 days, all of the formulations had released only 20 percent of the fluridone contained in the matrix. Further testing will be needed to test for release properties past 1 week. Laboratory CET studies indicate that fluridone concentrations of 10 to 50 ppb may have to be maintained for 28 to 70 days to achieve plant control (Hall, Westerdahl, and Stewart 1984; Netherland 1992).

The rate of water exchange will be a critical factor in determining the feasibility of using a fluridone CR matrix. High rates of water exchange could result in a slow-releasing CR matrix never reaching a lethal concentration in the treatment area, whereas a faster releasing CR matrix may not provide an adequate fluridone exposure period in the treatment area. Further studies concerning fluridone CET requirements for hydrilla and Eurasian watermilfoil will give us a better understanding of the most efficient way to apply controlled-release strategies or new delivery techniques when using fluridone.

The gypsum matrix formulated with triclopyr and 2,4-D showed good release characteristics for up to 6 days, at which point

Table 2
Fluridone Release Rates from Three CR Matrices

Herbicide	Matrix	Concentration, ppb							Percent Herbicide Remaining in Matrix
		D1	D2	D3	D4	D5	D6	D7	
Fluridone	Gypsum	<5	9	7	5	6	10	7	83
Fluridone	Protein	5.2	5	5	5	5	<5	<5	88
Fluridone	SRP	17	13	16	19	—	—	—	78

Table 3
2,4-D and Triclopyr Release Rates from CR Matrices

Herbicide	Matrix	Concentration, ppb							Percent Herbicide Remaining in Matrix
		D1	D2	D3	D4	D5	D6	D7	
2,4-D	Gypsum	0.11	0.20	0.22	0.15	0.21	0.19	0.08	86
2,4-D	Protein	0.40	0.50	0.24	0.14	0.05	0.03	0.03	70
2,4-D	BEE granular	0.73	0.56	0.12	0.14	—	—	—	77
Triclopyr	Gypsum	0.58	0.35	0.34	0.23	0.18	0.17	0.07	74
Triclopyr	Protein	1.2	0.18	0.09	0.07	<0.01	0.04	0.03	77

release rates began to drop off considerably. The protein matrix and the conventional 2,4-D granule resulted in higher concentrations on days 1 and 2, followed by a sharp drop in concentration by days 3 and 4. The drop-off in release rates was somewhat surprising, as approximately 80 percent of the herbicide was calculated to remain in all of the matrices. Triclopyr and 2,4-D have very similar CET requirements for the control of Eurasian watermilfoil (Green and Westerdahl 1990; Netherland and Getsinger 1992), and unlike fluridone, these compounds require only 1 to 3 days of exposure time at concentrations of 2.5 to 0.25 ppm. CR matrices that provide herbicide release for 2 to 10 days may be desirable for these products depending on water exchange characteristics.

Studies are being initiated to determine if greater efficacy is achieved by maintaining a constant exposure over time (constant release rate) or if an initial spike release followed by lower concentrations over time is more effective.

Future Work

Future work will include the testing of triclopyr slow-release matrices and of various delivery techniques in the Tennessee Valley Authority flume system. Residue analyses for bensulfuron methyl and endothall will be completed, and work on the internal tissue burden of herbicides will be initiated. The effect of increased loading rates on slow-release properties will also be evaluated.

Acknowledgments

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Field Testing of a Slow-Release Matrix Device (SRMD) in Flowing Water

by
David Sisneros¹

Introduction

Controlled-release (CR) herbicide technology provides several advantages over conventional types of applications, especially in flowing water. CR systems can increase the longevity of herbicide exposure (often necessary in areas of high water exchange), promote economic savings, reduce the number of herbicide treatments, and target specific areas or be manipulated to obtain the most effective coverage (Trimnell et al. 1982).

The Bureau of Reclamation and the US Army Corps of Engineers began a joint study in 1990 to evaluate a patented matrix currently marketed by Controlled Release Systems Research (CRSR). For test purposes, Rhodamine WT dye was substituted for an aquatic herbicide to facilitate onsite detection. The main objectives of this study were to determine if the matrix could provide a specific 14-day release rate in flowing water, to determine if the matrix could maintain a specified dye concentration of 10 ppb over a 14-day period in flowing water, to examine the dye concentration/exposure relationship in dense weedbeds, and to investigate flowing-water dynamics through dense noxious weedbeds.

This information is useful to understand the following factors: (a) water flow velocities and exchange rates in dense submersed plants stands, (b) maximum contact time of future herbicides utilizing the SRMD to provide reduced concentrations for prolonged periods, (c) offsite drift from the SRMD, and (d) simulated behavior of selected herbicides when applied to submersed plants.

Materials and Methods

SMRD design

Thirty-three SRMDs were obtained from CCSR. The prototype configuration and specifications are shown in Figure 1. The Rhodamine WT dye was approved by the Washington State Department of Ecology and is recommended by the US Geological Survey for typical water-tracing studies and has been approved for use in potable water at concentrations of up to 10 ppb.

Upper and lower plot design

Upper plot. An experimental plot was established in the Pend Oreille River north of Cusick, WA. This 0.4-ha midchannel plot (Figure 2) was 64 m on each side, and four sampling sites were established at the center of each quadrant within the plot. Additional sampling sites were located 61 m downstream of the lower edge of the plot and 30.5 m west of the plot. The average plot depth was 1.8 m. Flow rates were measured prior to testing at sampling sites within the plot and at corner markers at middepth using a March McBirney digital flowmeter. The average flow rate was 3.04 cm/sec. Flow rates in the Pend Oreille River ranged from 285,931 to 447,298 L/sec during this study.

Initially, 13 SRMDs were suspended at middepth approximately 15.2 m upstream of the leading edge of the plot and spaced at uniform intervals. Two additional SRMDs were installed—at 3 days and at 4 days post-treatment.

¹ US Bureau of Reclamation, Denver, CO.

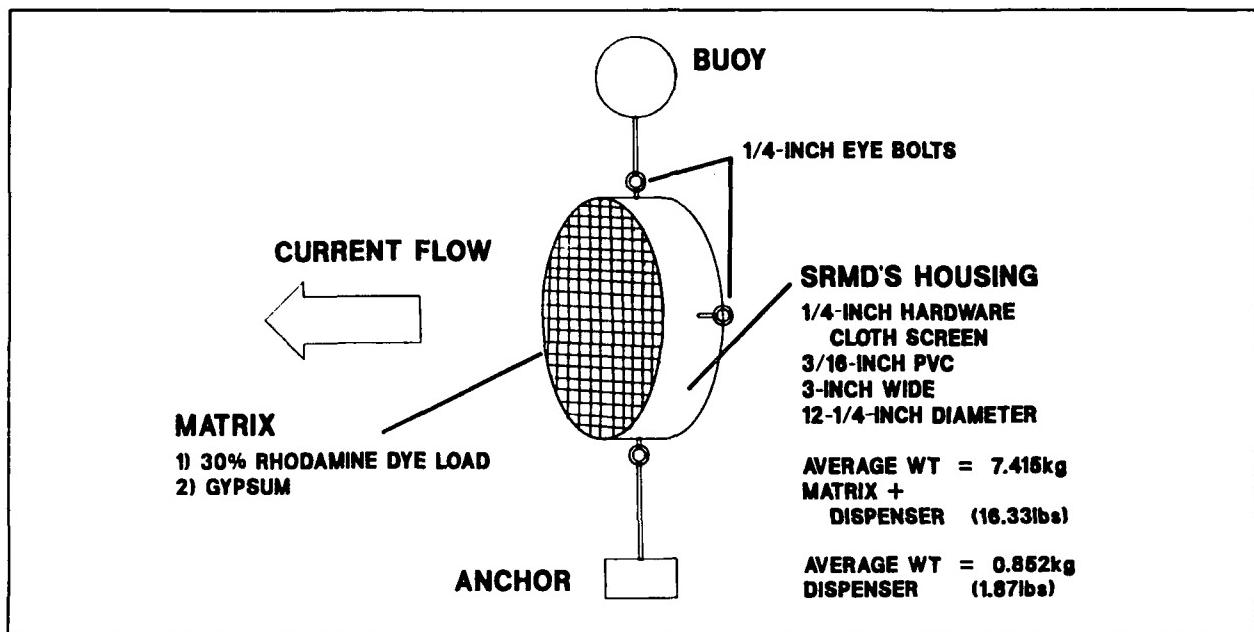


Figure 1. Prototype configuration of SRMD

Myriophyllum spicatum L. represented approximately 90 percent of the aquatic plant coverage within the plot and was within 15.2 cm of the water surface. The highest flow rate (6.1 cm/sec) was located at the

southeastern corner of the plot, adjacent to emergent vegetation which created the currents (marked by arrows in Figure 2). Dye concentrations were measured on a daily basis for 10 days.

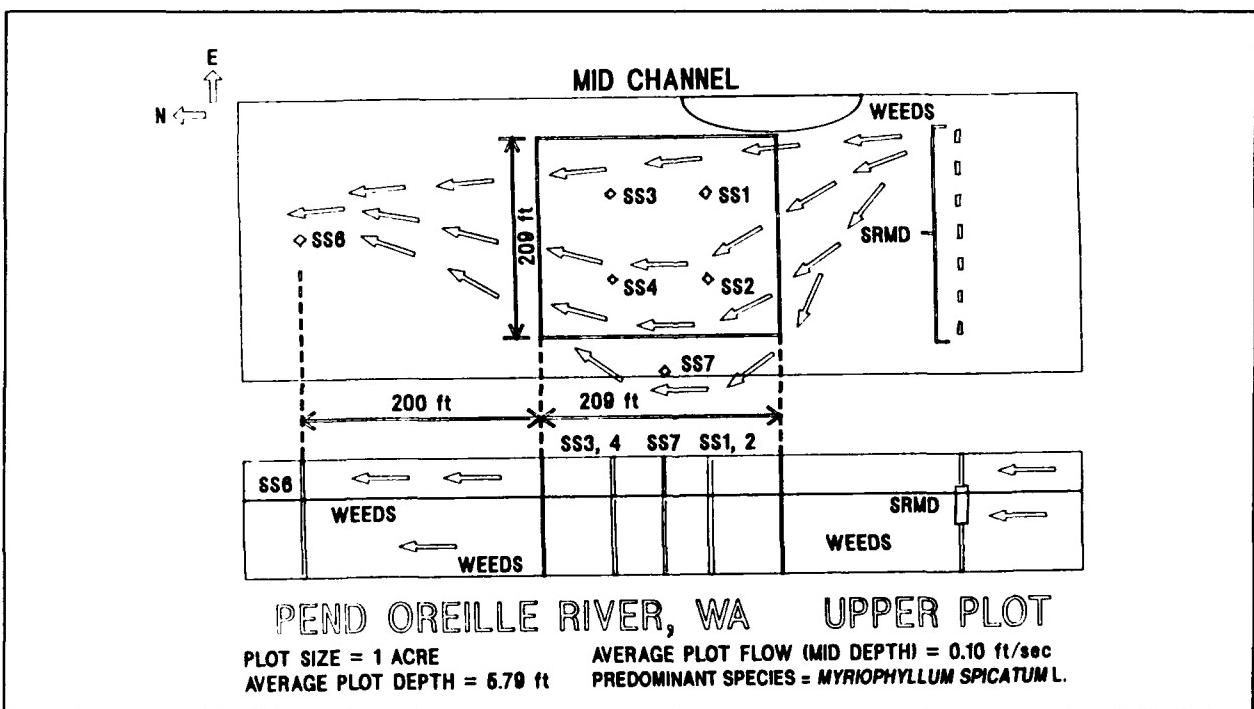


Figure 2. Design of upper plot, Pend Oreille River

Lower plot. The 1.09-ha plot (Figure 3) was located approximately 2.4 km downstream of the upper plot and was isolated from the main river channel by a small broken levee. This plot was 91.4 by 121.9 m and was marked using the same methods as for the upper plot. Five sampling sites were established within the plot. In addition, one sampling site was located downstream of the lower edge of the plot. The average plot depth was 1.8 m. Flow rates were measured in the same manner as in the upper plot, with an average flow of 2.1 cm/sec.

Initially, 13 SRMDs were installed, followed by one SRMD at 2 days posttreatment and two SRMDs at 3 days posttreatment. As with the upper plot, *Myriophyllum spicatum* L. covered 90 percent of the area within the plot. Dye concentrations were measured daily for 9 days, although no dye concentrations were measured at the 6-hr posttreatment sampling interval as in the upper plot.

Instrumentation

Dye concentrations were measured daily at each sampling site at 0.3-m intervals from the surface to the bottom of the water column. A Turner fluorometer equipped with a high-volume, continuous-flow cuvette, a chlorophyll-A plus Rhodamine dye filter kit, and a Fluke 52 K/J digital thermometer were used for determining dye concentrations. Water to be measured was circulated through the fluorometer with a Little Giant water pump attached to a 1.6-cm garden hose. Because the fluorescence of Rhodamine WT decreases with increased temperature, a correction for temperature with fluorometer calibration was necessary to determine actual dye concentration (Smart and Laidlaw 1977).

In addition, water quality parameters such as pH, dissolved oxygen (mg/L), conductivity (μS), oxidation/reduction potential (mV), and temperature ($^{\circ}\text{C}$) were measured daily.

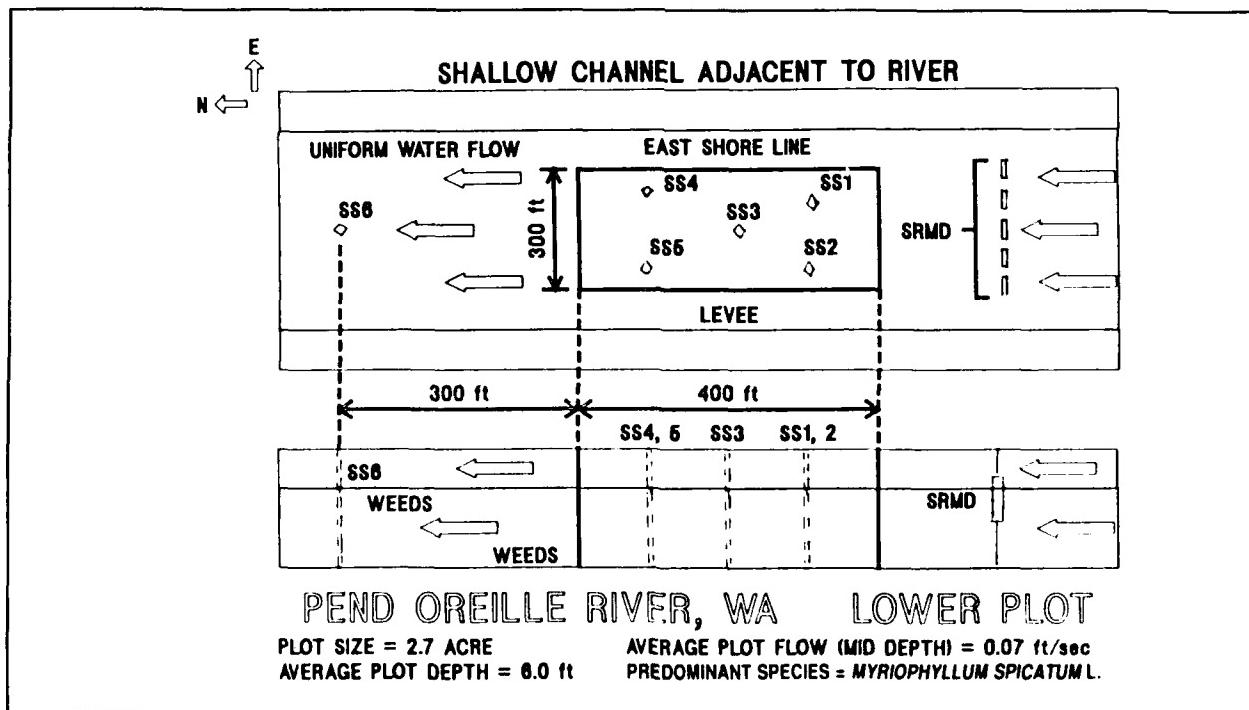


Figure 3. Design of lower plot, Pend Oreille River

These values were taken at one sampling site within each plot and one sampling site outside each plot at 0.3-, 0.9-, and 1.5-m depths using a Hydrolab Surveyor II multiparameter probe. Each SRMD was inspected daily to determine matrix consistency and longevity.

Results

Upper plot

Mean daily dye readings from sample sites were plotted over 10 days (Figure 4). Dye readings at outside sampling sites were not averaged. One proposed sampling site outside this plot was not established since access on the east of this plot was substantially reduced by shallow water depths and intense *Myriophyllum spicatum* growth. A representative dye concentration for the upper plot is shown in Figure 4. The specified dye concentration of 10 ppb is shown as a horizontal line parallel to the x-axis.

Initially, from 1 to 2 days postinstallation of the SRMD, there was a large release of dye followed by stepwise decline up to 10 days. In most cases, higher dye concentrations were found deeper in the water column, especially at 1.2, 1.5, and 1.8 m.

Dye concentrations at 0.3-, 0.6-, and 0.9-m depths more closely approximated the target release rate. In most cases, the target release rate of 10 ppb was met or exceeded throughout the water column within the plot. Dye concentrations were found approximately 61 m downstream from the lower edge of the plot for up to 7 days. Some lateral movement of dye was seen west of the plot up to 3 days; however, dye concentration declined from 4 to 10 days.

Lower plot

As with the upper plot, daily mean dye concentrations were plotted over 9 days (Figure 5). Dye concentrations outside the plot

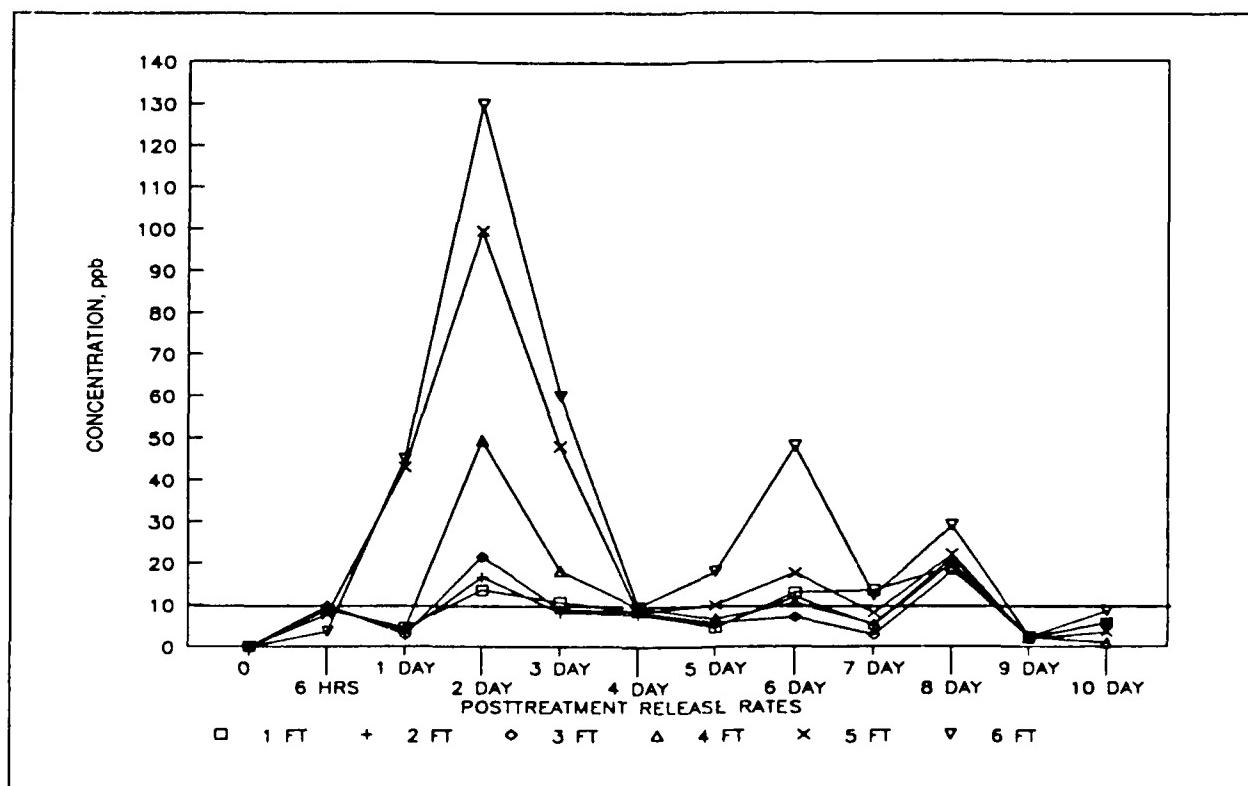


Figure 4. Rhodamine WT dye concentration at sampling sites S1 and S2, upper plot

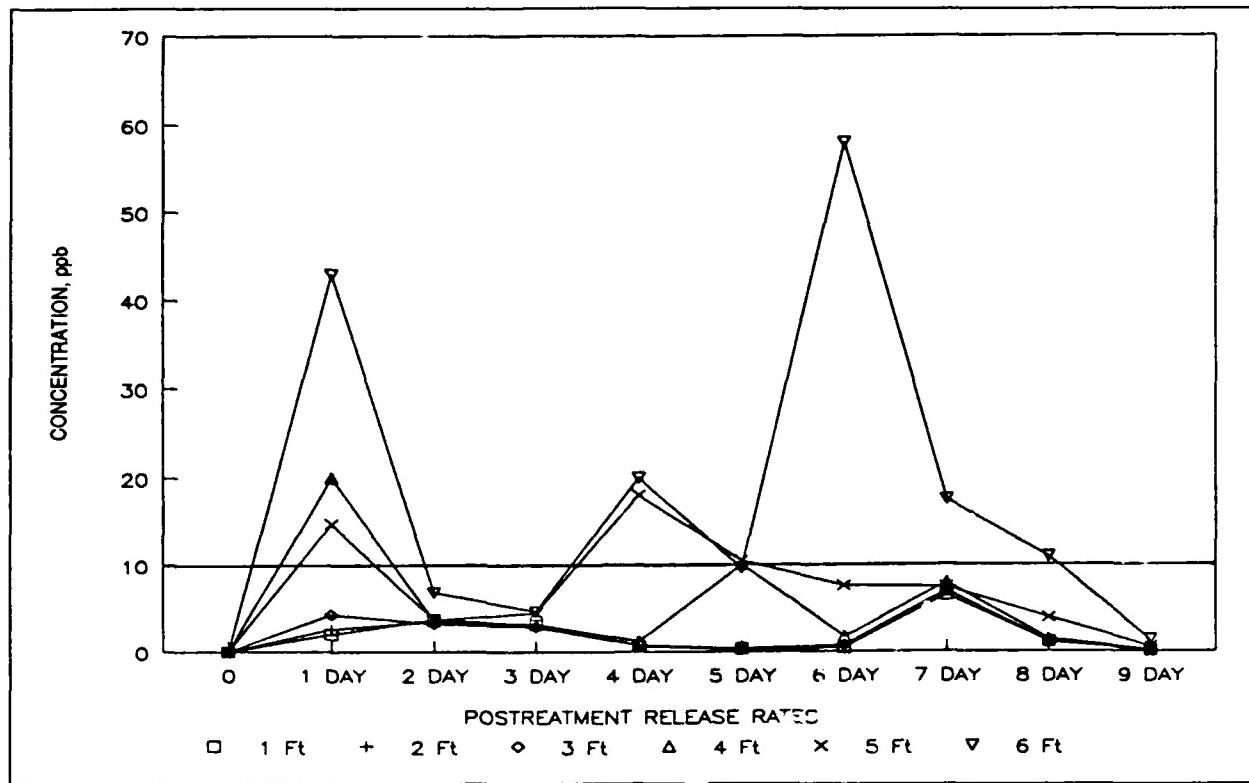


Figure 5. Rhodamine WT dye concentration at sampling sites S1 and S2, lower plot

were not averaged. Initially, at the 1-day postinstallation sampling interval, a large release of dye was detected at all points as in the upper plot. The specified dye concentration of 10 ppb is shown as a horizontal line parallel to the x-axis. At a majority of the sampling intervals, higher dye concentrations were in the water column, especially at 1.2, 1.5, and 1.8 m. These values met and exceeded the target release rate of 10 ppb. Dye concentration at sampling depths of 0.3, 0.6, and 0.9 m more closely approximated the target release rate than the lower depths.

During a number of sampling intervals in the higher portions of the water column (0.3, 0.6, and 0.9 m), there was a general decline in dye concentration from 3 to 6 days post-installation followed by slight rises thereafter. These decreases could have been due to increases in river flows which were at their highest on August 10, 1990, corresponding to the 5-day postinstallation sampling interval. In addition, there was an increase in dye concentration at the 6-day sampling interval at the upper sampling stations.

Measurable dye concentrations were found at most sampling depths and sampling intervals. A decline at the downstream sampling site at 5 days was possibly attributed to dilution by the high flow rate of 447,298 L/sec.

Conclusions

Overall, the SRMDs did not provide the specified 14-day release rate. Longevity of the SRMDs ranged from 3 to 8 days, which was likely the result of the low concentration of dye used to make the matrix workable. As a result, instead of the matrix expanding to fit against the housing, the matrix shrank away, leaving large gaps that increased the exposed surface area and may have resulted in decreased longevity. In addition, the matrix is usually hard; however, because of the amount of water needed to make the matrix plus dye workable, this particular matrix was soft and less resistant to erosion by water flow, possibly giving the reduced longevity.

A dye concentration of 10 ppb was not maintained for 14 days in both plots. However, dye concentration of 10 ppb was maintained or exceeded in sampling sites within the upper and lower plot, especially in the lower sampling depths for 10 and 9 days, respectively.

In the upper plot, channeling occurred and was especially visible toward the upper end of the plot as distinct colored bands. Toward the lower end of the plot, channeling was not evident. Individual SRMDs that were installed at the upper plot at 3 and 4 days posttreatment were placed outside the southeastern portion adjacent to the emergent vegetation. This resulted in better coverage to the eastern portion of that plot. Observations of the lower plot during sampling indicated that homogeneous mixing had occurred throughout.

Potentially, the SRMDs could be used to encapsulate low-rate herbicides such as bensulfuron methyl (Mariner) and triclopyr (Garlon) for the control of noxious aquatic plants. These are herbicides that have been evaluated in field dissipation trials at Banks

Lake, Washington, and have shown excellent efficacy in other trials.

Acknowledgments

The author expresses appreciation for the technical assistance and support provided to this study by the US Army Engineer District, Seattle; the Libby-Albeni Falls Project Office; the Washington Department of Ecology; Aquatics Unlimited; and the WES. Appreciation is also extended to Ms. Stephanie Smink, US Bureau of Reclamation, for field assistance.

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Controlling Submersed Plants with Herbicides in Flowing Water Systems

by

Kurt D. Getsinger,¹ Alison M. Fox,² and William T. Haller²

Introduction

The successful control of submersed aquatic plants using chemicals depends upon the concentration and exposure time of a herbicide with respect to the target plant. In high water-exchange systems (e.g. rivers, tidal areas, and large reservoirs), the movement of water can dramatically impact herbicide concentration/exposure time (CET) relationships, resulting in reduced chemical contact time and efficacy. In an effort to improve the control of nuisance species (such as Eurasian watermilfoil and hydrilla) in flowing water, researchers at the US Army Engineer Waterways Experiment Station (WES) and the University of Florida Center for Aquatic Plants are evaluating conventional and innovative submersed application techniques in selected high-water exchange systems around the Nation (Getsinger, Green, and Westerdahl 1990; Getsinger et al. 1991). This article provides an update on herbicide application technique development for flowing water.

Approach

One key to improving chemical control of submersed plants in dynamic situations is to

develop an understanding of water movement within the plant stands of a specific flowing-water system. Estimates of water movement within plant stands can be determined using flowmeters, tracer dyes, weirs, and gaging stations. Once obtained, this information can be analyzed to predict herbicide contact time around the target plants. The second critical piece of information is to select the appropriate herbicide, based on plant susceptibility, estimated herbicide contact time, and specific herbicide CET relationships. This approach has been successfully demonstrated in five flowing-water systems, using three herbicides against two target plants (Table 1). Treatment strategies and efficacy results for each of these demonstrations are summarized below.

Treatment Strategies and Results

Crystal River

Spring-fed tidal canals in Crystal River, Florida, have been heavily infested with hydrilla for over 20 years. This infestation has required extensive management by chemical and mechanical methods to maintain navigation for recreational boating; however, the efficacy of aquatic herbicides has been variable and unpredictable.

Table 1
Flowing-Water Systems, Target Plants, and Herbicides Selected for Evaluation of Submersed Application Techniques

Flowing Systems	Target Plant	Herbicide
Crystal River, Florida	Hydrilla	Endothall
St. Johns River, Florida	Hydrilla	Fluridone
Withlacoochee River, Florida	Hydrilla	Fluridone
Long Lake, Washington	Eurasian watermilfoil	Fluridone
Pend Oreille River, Washington	Eurasian watermilfoil	Triclopyr

¹ US Army Engineer Waterways Experiment Station, Vicksburg, MS.

² University of Florida Center for Aquatic Plants, Gainesville, FL.

In 1987-89, a series of dye studies were conducted to determine water exchange characteristics in the hydrilla-infested Three Sisters Canals (Fox, Haller, and Getsinger 1991). Data from these studies indicated that water temperature played a significant role in water movement patterns on a seasonal basis in Three Sisters Canals, and this information was used to develop chemical treatment strategies for the canal system. As a demonstration, the herbicide endothall was applied to the canals during the fall months, when water exchange patterns allowed for extended periods of chemical contact time within the hydrilla stands. This treatment strategy resulted in much improved, and predictable, herbicide efficacy compared to historical treatments.

Based on results from the dye studies and successful herbicide demonstrations, recommendations for chemical control of hydrilla in the Three Sisters Canals (and associated areas) were provided to the Jacksonville District and the Citrus County Aquatic Plant Management Program.

St. Johns and Withlacoochee Rivers

Extensive stretches of the relatively slow-moving upper St. Johns and Withlacoochee Rivers, Florida, have been infested with hydrilla for 10 to 15 years. As a result of the variable water flow that occurs in these systems, aquatic plant control efforts using herbicides have been inconsistent. Using river discharge data and information from dye studies, chemical treatment strategies were devised for controlling hydrilla in selected portions of the rivers (Fox and Haller 1990; Getsinger, Fox, and Haller 1990; Haller, Fox, and Schilling 1990).

Programs were developed to treat hydrilla with the herbicide fluridone in upstream portions of each river, relying upon water flow to mix and distribute the herbicide downstream to achieve a dose of 10 to 15 µg/L (considerably below the US Environmental Protection Agency's (EPA) water tolerance for fluridone of 150 µg/L) for up to 12 weeks. The low herbicide doses and long contact times used in these treatment programs (confirmed via

water residue analyses) were based on fluridone CET relationships being developed in concurrent laboratory and field studies.

Implementation of these treatments resulted in excellent control of hydrilla in a 16-km stretch of the St. Johns River and a 29-km stretch of the Withlacoochee River. Recommendations for hydrilla control strategies in the St. Johns and Withlacoochee Rivers using chemicals are being provided to the Jacksonville District and to the St. Johns and Southwest Florida Water Management Districts.

Long Lake

In 1987, Long Lake (a 135-ha reservoir near Olympia, WA) was invaded by Eurasian watermilfoil. By 1990, milfoil had occupied more than two thirds of the lake's surface area, while also infesting several smaller, downstream lakes. Using lake water retention time data and laboratory-derived herbicide CET relationships, a treatment strategy was designed to maintain fluridone levels in the lake at 20 to 40 µg/L (well below the EPA water tolerance for fluridone of 150 µg/L) for 8 to 12 weeks. A series of four fluridone treatments were made from 2 July to 14 August 1991. These treatments were applied to different 2-to 4-ha sites around the lake, with application sites being concentrated in the upstream portion of the reservoir. This allowed the slow water movement to distribute the herbicide throughout the system, thereby achieving the desired fluridone concentrations and exposure times (confirmed via water residue analyses).

The fluridone application provided excellent control of milfoil, with minimal to no injury symptoms apparent on nonweedy shoreline and associated wetland vegetation. A 1-year posttreatment assessment of plant control and nontarget impacts will be conducted in fiscal year (FY) 1992.

Pend Oreille River

The herbicide triclopyr was successfully used to control Eurasian watermilfoil in riverine and cove sites on the Pend Oreille River

in eastern Washington in August 1991. An update on this study is presented in a separate article in this proceedings (Getsinger, Turner, and Madsen 1992).

Future Work

Research in FY 92 will continue to focus on improving the control of submersed plants in high water-exchange environments through the development and evaluation of application techniques. Field studies are scheduled for Alabama, Florida, and the Pacific Northwest.

Acknowledgments

The authors express appreciation for the technical assistance and support provided by the Jacksonville and Seattle Districts, Washington State Department of Ecology, Thurston County Public Works, Long Lake Steering Committee, Citrus County Aquatic Plant Management Program, St. Johns and Southwest Florida Water Management Districts, DowElanco, Atochem, and numerous personnel from the WES Chemical Control Technology Team and the University of Florida Center for Aquatic Plants.

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Field Evaluation of the Herbicide Triclopyr

by

Kurt D. Getsinger,¹ E. Glenn Turner,¹ and John D. Madsen¹

Introduction

The herbicide triclopyr (3,5,6-trichloro-2-pyridinyloxyacetic acid), formulated as a triethylamine salt (Garlon 3A), has been used for over 15 years to control broadleaf weeds in forestry, industrial, and other noncrop terrestrial sites. Triclopyr is an auxin-like, systemic herbicide with a mode of action and spectrum of weed control similar to that of phenoxy herbicides, such as 2,4-D (2,4-dichlorophenoxy acetic acid). Manufactured by DowElanco, triclopyr is registered for aquatic sites through 1992 under a Federal Experimental Use Permit (EUP). The US Environmental Protection Agency is currently considering the product for full aquatic registration.

Previous field evaluations have shown that triclopyr can provide aquatic plant managers with a feasible alternative to 2,4-D for controlling Eurasian watermilfoil, waterhyacinth, alligatorweed, melaleuca, purple loosestrife, and other nuisance vegetation (Getsinger and Westerdahl 1984, Langeland 1986, Green et al. 1989, Wujek 1990).

Results from concentration/exposure time (CET) studies conducted at the US Army Engineer Waterways Experiment Station (WES) showed that triclopyr provided excellent control of the submersed species Eurasian watermilfoil (hereafter called milfoil) under laboratory conditions when that plant was exposed to concentrations ranging from 2.5 to 0.25 mg acid equivalent (ae)/L triclopyr for 18 to 72 hr (Netherland and Getsinger 1992).

In an effort to verify results from these laboratory studies, and to evaluate the species-selective properties of triclopyr, WES

researchers have been applying triclopyr under an EUP to milfoil-dominated plant communities in the Pend Oreille River, Washington; Guntersville Reservoir, Alabama; and other locations. Results from these field evaluations will be used to provide guidance for the use of triclopyr in aquatic systems. This article provides an update on the Pend Oreille River triclopyr field studies.

Materials and Methods

Two plots, located in the Pend Oreille River, were selected for triclopyr treatment. A 6-ha (15-acre) plot was established in a shallow region of the river near river mile (RM) 61, while a 4-ha acre plot was established in a protected cove near RM 48 (Figures 1 and 2). An additional area, located upstream from the herbicide applications, was selected to serve as an untreated reference plot (Figure 1).

Water depth in the plots ranged from 0.25 to 2.8 m, and all plots contained dense populations of milfoil, associated with lesser amounts of native macrophytes (e.g. elodea, coontail, and pondweeds). In deeper areas of the plots, tips of milfoil plants were 15 to 20 cm from the water surface; however, in most areas less than 1.5 m deep, a dense milfoil surface mat had developed.

A tank mix of Garlon 3A and Rhodamine WT (RWT) dye was evenly applied to the river and cove plots, using an airboat, on the mornings of 21 and 22 August 1991, respectively. This mixture was applied at rates calculated to achieve the following concentrations:

¹ US Army Engineer Waterways Experiment Station, Vicksburg, MS.

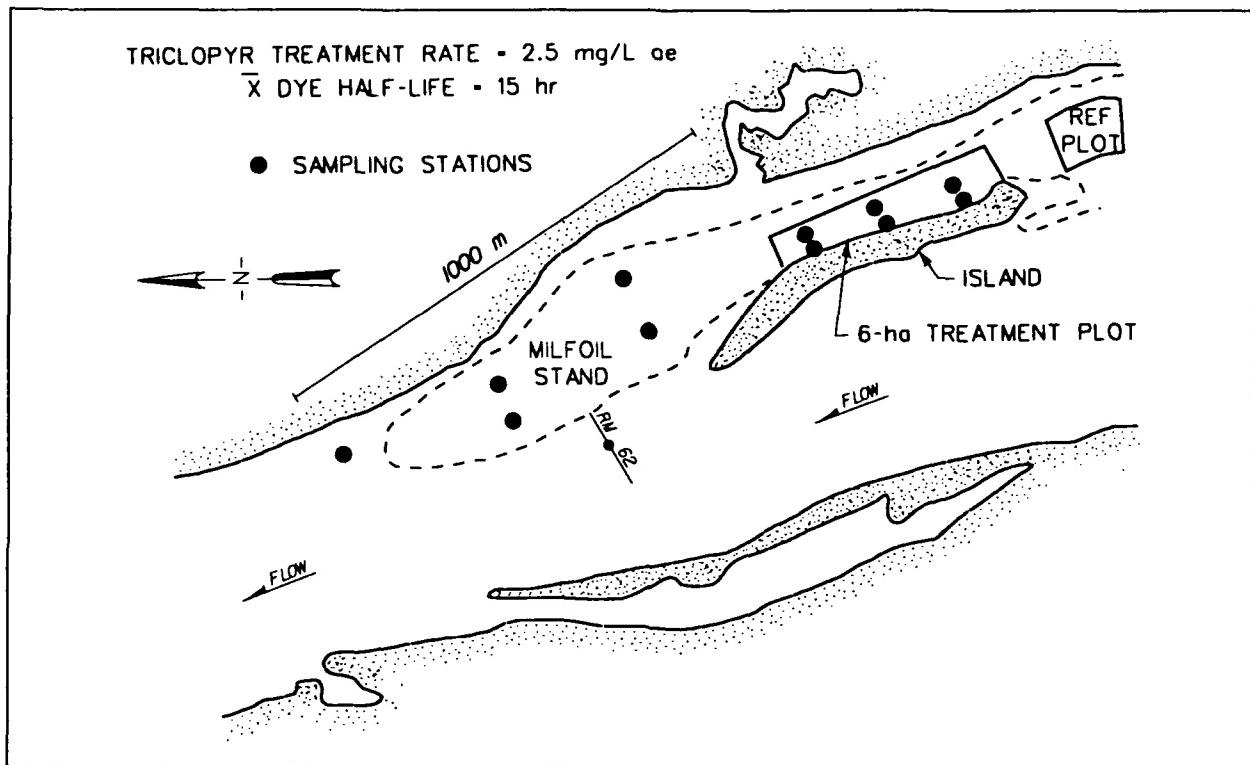


Figure 1. Pend Oreille River dye/triclopyr treatment and reference plots, August 1991

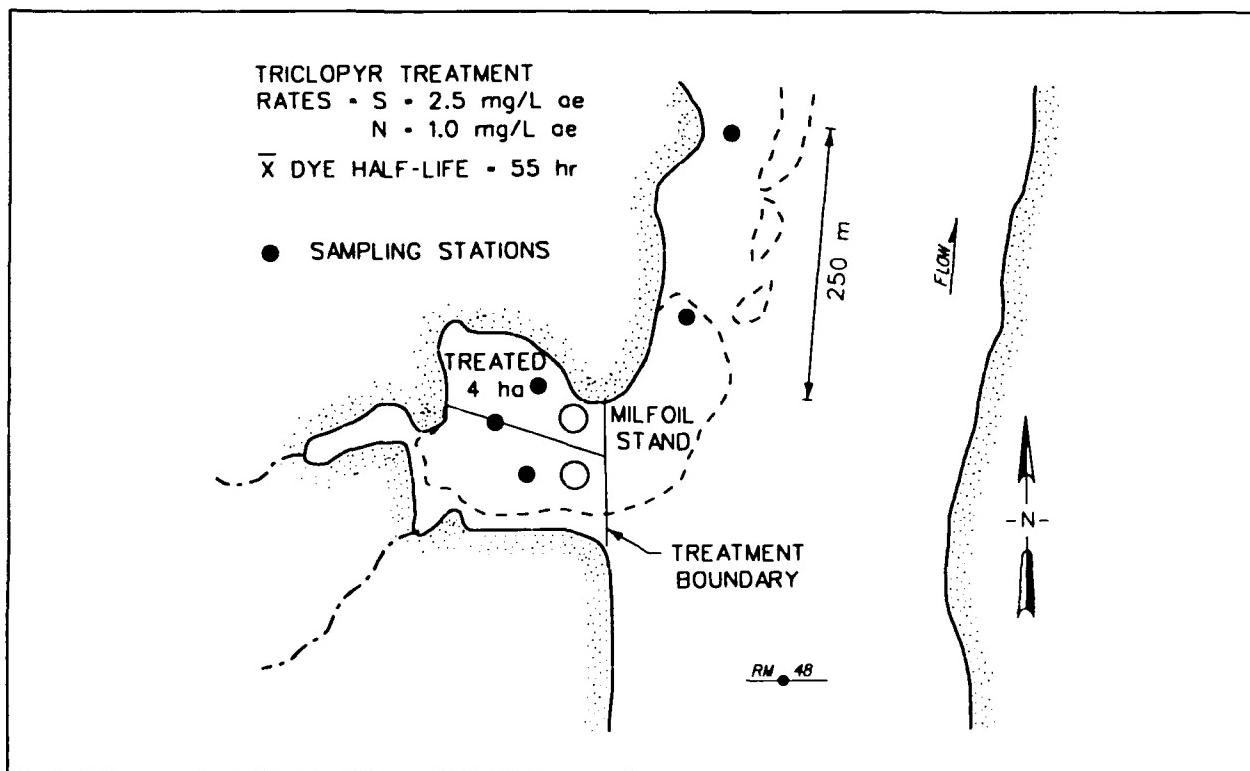


Figure 2. Lost Creek Cove dye/triclopyr treatment plot, August 1991

- In the river plot, 2.5 mg ae/L (ppm) triclopyr + 10 µg/L (ppb) RWT.
- In the southern half of the cove plot, 2.5 mg ae/L triclopyr + 10 µg/L RWT.
- In the northern half of the cove plot, 1.0 mg ae/L triclopyr + 4 µg/L RWT.

These treatment rates were selected based on information obtained from a previous water-exchange study conducted in the plots in August 1990 (Getsinger et al. 1991). Results from that 1990 dye study showed that water-exchange half-lives averaged approximately 8 hr in the river plot and 40 hr in the cove plot (with a 17/73 hr, south/north half-life split in the cove plot). This water-exchange information allowed for a 30-percent reduction in the amount of Garlon 3A to be applied to the cove (compared to the maximum amount permitted under the Garlon 3A EUP label).

Posttreatment dye concentrations were monitored at selected sampling stations within and outside the plots. Instantaneous dye readings taken during the study were used to predict any off-target herbicide movement, thereby improving the selection of water sampling stations located outside the treated plots. In addition, water samples were concurrently collected with these dye measurements and stored for later triclopyr residue analysis. These data will be used to compare the dissipation of triclopyr with that of the dye following application. To characterize herbicide efficacy and selectivity, pretreatment and posttreatment plant diversity and biomass samples were collected in all plots.

Results and Discussion

Analysis of posttreatment data and observations indicated that triclopyr provided excellent control of milfoil in both the river and cove plots. Milfoil shoots displayed epinastic twisting by 24 hr after herbicide application, and began to sink to the bottom within 3 days posttreatment. At 1 week posttreatment, all milfoil shoots in the treated plots lay prostrate on the sediment, with leaves browning and dropping from the stems. During this period of milfoil degradation, some understory native plants were trapped by the sinking milfoil mats and pulled to the bottom. This situation undoubtedly contributed to a partial decline of native plants in the treated plots. Milfoil in the untreated reference plot remained vigorous and healthy.

The 4-week posttreatment biomass data showed a 99-percent reduction in milfoil shoot mass in both treated plots (Table 1). Although native plant biomass was reduced by 54 percent in the river plot and 76 percent in the cove plot, a similar reduction in native shoot mass (58 percent) was measured in the untreated reference plot. Some of this reduction in native plant biomass can be attributed to late-season senescence of these plants, as winter bud formation was observed on the pondweeds in all plots.

A visual evaluation at 8 weeks posttreatment indicated that milfoil shoots had completely decomposed in both treated plots, but were still upright and green in the untreated reference area. Furthermore, scattered patches of short native shoots (primarily elodea and coontail) were observed throughout the treated plots. Plant biomass and species

Table 1
Mean Pretreatment and 4-Week Posttreatment Shoot Mass for Garlon 3A-Treated Plots,
Pend Oreille River, 1991

Plot	Eurasian Watermilfoil		Native Plants	
	Pretreatment	4-Week Posttreatment	Pretreatment	4-Week Posttreatment
Reference	291 (± 48) ¹	281 (± 36)	12 (± 6)	5 (± 3)
River	254 (± 46)	3 (± 2)	39 (± 20)	18 (± 10)
Cove	257 (± 39)	2 (± 1)	38 (± 16)	9 (± 4)

¹ Values are in grams dry weight per square meter (± 1 standard error).

diversity assessments will be conducted at 1 year posttreatment to determine the extent of milfoil control and recolonization.

Dye dissipation half-lives (Table 2) characterized water exchange (and potential herbicide dissipation) within the treated plots. This water-exchange information can be used to calculate theoretical herbicide half-lives within the plots. When these triclopyr half-life values are plotted against laboratory-derived triclopyr CET relationships, milfoil control in the field can be estimated. Figure 3 shows that sufficient triclopyr contact time was maintained to provide excellent initial knock-down and acceptable control of milfoil in the Pend Oreille treatments (verified at the 4-week posttreatment biomass evaluation). Moreover, these exposure periods should provide large areas of complete milfoil rootcrown destruction, particularly in the cove plot.

A final report, summarizing pretreatment and posttreatment plant biomass, species di-

versity, triclopyr efficacy, and dye/triclopyr water dissipation relationships, will be published following the 1-year posttreatment evaluation.

Table 2
Mean Half-Lives of Dye Dissipation
for Rhodamine WT-Treated Plots,
Pend Oreille River, 1991

Plot	Half-Life, hr	r ²
River	15 ¹	99
Cove	55	91

¹ Values represent mean of all internal sampling stations within each plot.

Summary and Future Work

Preliminary results from the Pend Oreille River field study have demonstrated that triclopyr can be an effective herbicide for the selective control of milfoil, given sufficient contact time. The Pend Oreille cove treatment also demonstrated that if water-exchange

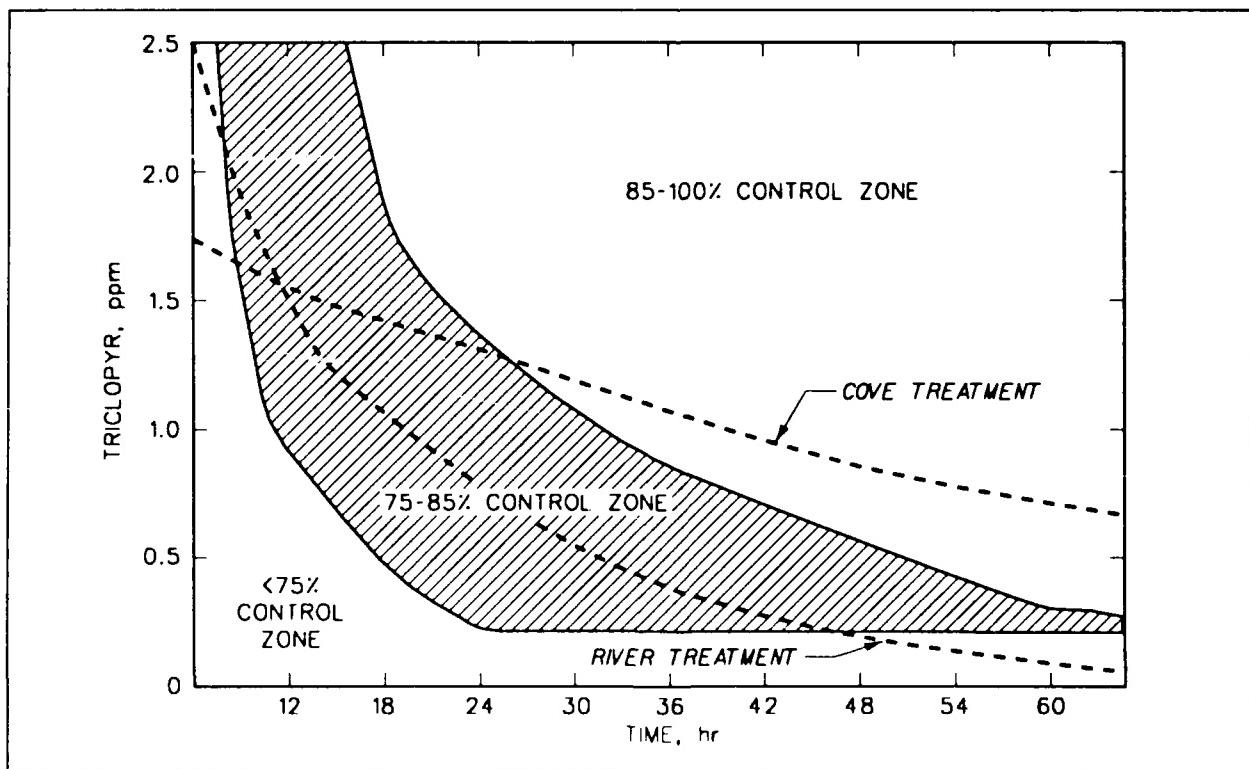


Figure 3. Predicted milfoil control on the Pend Oreille River using water-exchange information and laboratory-derived concentration/exposure time relationships

patterns can be determined within the target plant stand, herbicide doses below the maximum label rates can be used to provide acceptable plant control. Researchers at the WES will continue to evaluate triclopyr and other promising aquatic herbicides in the field, emphasizing the use of water-exchange information to improve the control of submersed plants in flowing-water systems.

Acknowledgments

Technical assistance and support for this study were provided by the Seattle District, the Libby-Albeni Falls Project Office, the Washington Department of Ecology, the Pend Oreille County Noxious Weed Control Board, Aquatics Unlimited, and DowElanco.

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Plant Growth Regulators for Aquatic Plant Management

by

Linda S. Nelson¹ and Michael D. Netherland¹

Introduction

Plants function as vital components of the aquatic ecosystem, providing oxygen through photosynthesis, food and habitat for aquatic organisms, as well as bottom sediment stabilization. On the contrary, many chemical methods available for managing nuisance levels of aquatic plants are designed to kill all, or most, of the vegetation in an area of treatment, significantly reducing the beneficial aspects that submersed plant communities provide. The use of plant growth regulators (PGRs) may offer a new, compromising alternative to traditional chemical management techniques, allowing plants to remain as a part of the aquatic ecosystem by limiting or suppressing their growth to nonnuisance levels.

PGRs are synthetic compounds that, when applied to plants in small amounts, promote, inhibit, or modify plant growth and/or development. Modifications may include improved yield, enhanced quality, fruit abscission, and reduced stem elongation. Many herbicides also exhibit growth-regulating effects when applied at sublethal concentrations (Klaine and Knowles 1988). To date, PGRs have been exclusively marketed for use on terrestrial plants; however, recent studies demonstrating their effectiveness on aquatic plant species (Van 1988, Lembi and Netherland 1990, and Kane and Gilman 1991) have stimulated industry interest in the aquatics market.

The objectives of this work unit are to evaluate PGR activity on the growth and reproduction of nuisance aquatic plant species and to determine the feasibility of using PGRs as a management strategy. Cooperative research

is currently being conducted by Purdue University, the USDA Aquatic Plant Management Laboratory at Fort Lauderdale, and the US Army Engineer Waterways Experiment Station (WES). Research updates from each of the performing agencies are summarized in this proceedings.

Studies at the WES are directed toward evaluation of the compound bensulfuron methyl and its activity on Eurasian water-milfoil (*Myriophyllum spicatum* L.). Bensulfuron methyl is one of the newest and most promising compounds for use in aquatic systems. This compound is a member of the sulfonylurea herbicide group developed by E. I. Du Pont de Nemours & Company. Sulfonylureas, in general, are known to have high levels of activity at very low application rates. Their mode of action is inhibition of the plant enzyme acetolactate synthase, which is necessary for amino acid synthesis (Beyer et al. 1988). Inhibition of this enzyme thus interrupts or slows plant growth. Several sulfonylureas are currently marketed for use in agriculture; in fact, bensulfuron methyl was first introduced and is fully registered under the trade name Londax, as a broadleaf and grass herbicide for use in rice production. Bensulfuron methyl also shows potential as a herbicide and growth regulator (when used at lower rates) for management of submersed aquatic plants (Anderson and Dechoretz 1988, Lembi and Netherland 1990). Investigations with this compound in aquatic systems are currently conducted under an Experimental Use Permit issued by the US Environmental Protection Agency (EPA). When the compound receives full registration by EPA, the aquatic formulation will be marketed by Du Pont under the trade name Mariner.

¹ US Army Engineer Waterways Experiment Station, Vicksburg, MS.

Materials and Methods

Experiments were conducted at the WES in a controlled-environment aquarium system. The system consisted of twenty-four 55-L aquaria (0.75×0.3 m), each independently supplied with a continuous flow of reconstituted hard water that allowed the total water volume of each aquarium to be exchanged every 24 hr. Air was bubbled through each aquarium to enhance water circulation. Water temperature was maintained at 25 ± 2 °C throughout the experiment. Overhead supplemental lighting provided a light:dark cycle of 13:11 hr, with a mean photosynthetically active radiation at the water surface of $450 \mu\text{E}/\text{m}^2$.

Eurasian watermilfoil used in this study was supplied by the WES Lewisville Aquatic Ecosystem Research Facility (LAERF), Lewisville, TX. Watermilfoil was separated into 10-cm apical segments and planted 5 cm deep into sediment-filled beakers. The sediment used was collected from Brown's Lake, Mississippi, and was amended with Rapid-gro to avoid any nutrient limitations.

Nine beakers were placed into each aquarium, and plant segments were allowed to establish new shoot and root growth. Following 2 weeks of growth, plants were trimmed to a height of 20 cm; 1 week thereafter, chemical treatments were applied. One beaker of plants was randomly removed from each aquarium immediately prior to treatment to provide an estimate of treated biomass.

Established plants were exposed to static (continuous flow-through water system turned off) treatments of varying bensulfuron methyl concentrations for 14-, 21-, and 28-day time periods (Table 1). Following the exposure period, the aquaria were drained and rinsed three times to remove chemical-treated water, after which the continuous flow-through water system was resumed for the duration of the experiment.

Treatments were arranged in a completely randomized design with three replications.

Visual ratings of plant injury were recorded weekly. At the conclusion of the experiment (5 weeks posttreatment), root and shoot biomass were measured for each treatment. Data were analyzed using analysis of variance, and treatment effects were separated using the Waller-Duncan Test.

Table 1
Bensulfuron Methyl Treatment Rates and Exposure Time Periods

Rate, µg/L or ppb	Exposure Time, days
0 (Control)	0
50	14
75	14
5	21
10	21
25	21
50	21
5	28

Results

Five treatments significantly reduced watermilfoil shoot biomass (Figure 1). Reductions ranged from 26 to 69 percent when compared to untreated plants, with the most effective treatment being a 21-day exposure to 50 µg/L bensulfuron methyl. Regrowth was observed to some extent on all treatments by the end of the experiment, and plants had "topped out" or grown to the water surface on all but two treatments (25 and 50 µg/L at 21-day exposures). Regrowth emerged from rootcrown, lateral buds along stem nodes, and apical tips, and was evident 1 to 2 weeks following removal of the chemically treated water.

Although the data indicated significant reductions in biomass production, topped-out vegetation signifies strong regrowth potential and may be indicative of an inadequate treatment. Despite the slight increases in root biomass with several treatments, no significant differences in root growth were observed compared to untreated plants (Figure 2).

The results indicate that bensulfuron methyl is effective at reducing the growth of Eurasian watermilfoil, and support earlier

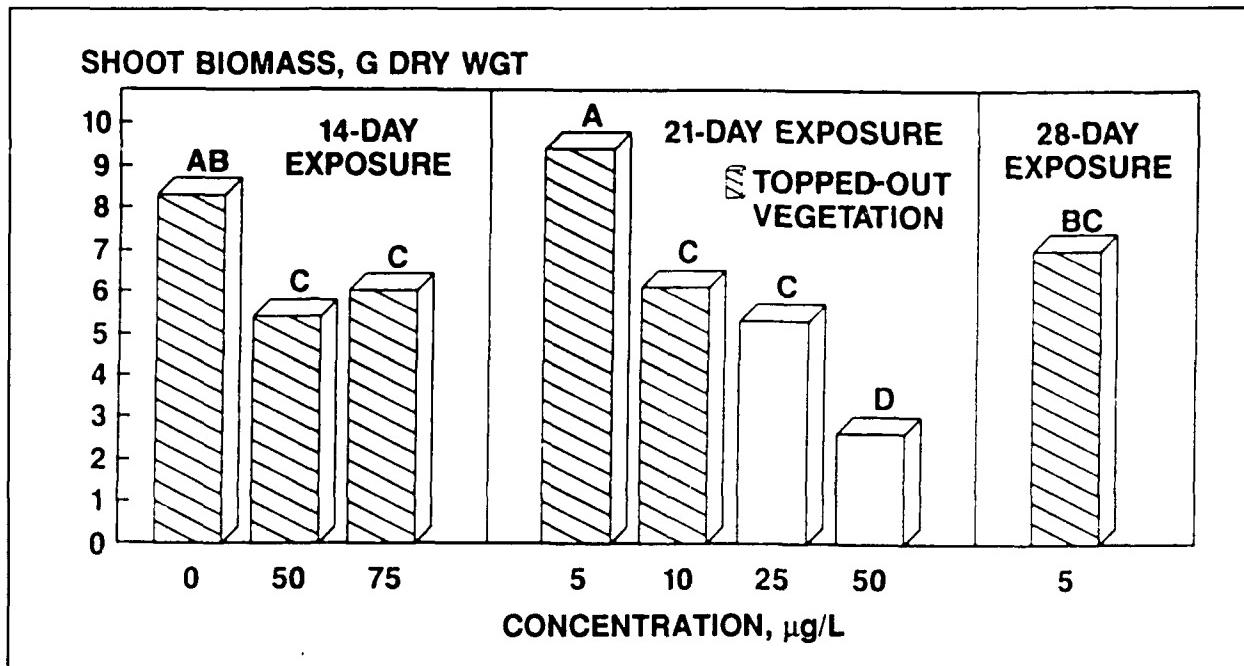


Figure 1. Effects of bensulfuron methyl on shoot biomass of Eurasian watermilfoil 5 weeks after treatment. Data are means of three observations. Letters denote significant differences at $P = 0.05$

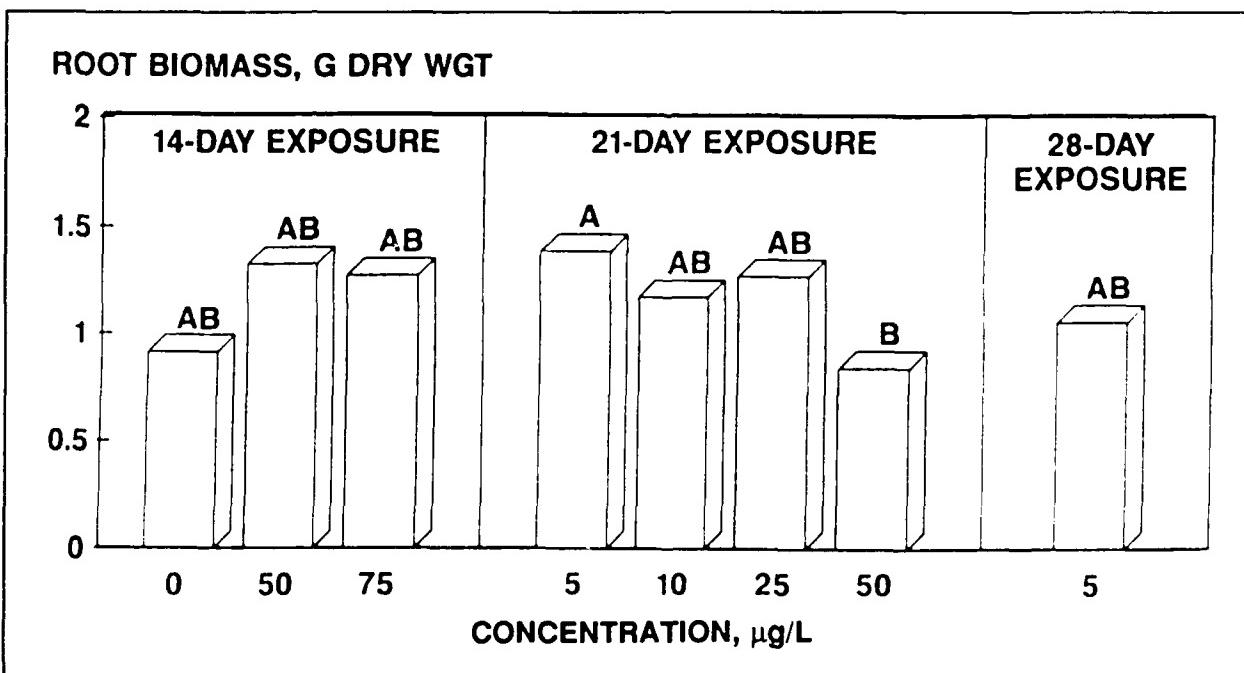


Figure 2. Effects of bensulfuron methyl on root biomass of Eurasian watermilfoil 5 weeks after treatment. Data are means of three observations. Letters denote significant differences at $P = 0.05$

conclusions by Anderson and Dechoretz (1988). Under these experimental conditions, a 21-day exposure period to concentrations of 25 to 50 µg/L was necessary to maintain acceptable growth suppression (not topped out). Higher concentrations at lower exposure periods were less effective, suggesting that contact time is an important factor in determining treatment success.

Future Work

Future studies to be conducted by the WES in this work unit include the following:

- Conduct mesocosm studies at the LAERF to validate laboratory results on bensulfuron methyl versus Eurasian watermilfoil.
- Initiate studies in the WES aquarium system to further evaluate flurprimidol versus hydrilla (*Hydrilla verticillata* (L.f.) Royle) and Eurasian watermilfoil. Initial results of laboratory bioassay tests indicate that flurprimidol is effective at significantly reducing stem length of Eurasian watermilfoil and hydrilla, without disrupting the physiological competence (photosynthesis and respiration) of these plants (Lebbi and Netherland 1990).

Acknowledgments

The authors would like to thank Mr. Glenn Turner, Dr. Susan Sprecher, and Mses. Anne Stewart and Sheron Burt for assistance in conducting this study.

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Evaluation of Bensulfuron Methyl for Hydrilla Control

by

Thai K. Van¹ and Vernon V. Vandiver, Jr.²

Introduction

One major problem encountered in hydrilla control is the rapid regrowth of the plant from vegetative propagules. The subterranean turions, commonly called tubers, are particularly troublesome since they serve as a source of regrowth in areas where the hydrilla plants have been controlled by chemical or mechanical methods. Dioecious hydrilla produces tubers in North Florida from October through April (Haller, Miller, and Garrard 1976) in response to short photoperiods (Van, Haller, and Garrard 1978). This seasonality of tuber production suggests that the timing of control procedures is important for effective hydrilla management.

More recent studies, however, have indicated that monoecious hydrilla produces tubers under both 10-hr and 16-hr photoperiods (Van 1989), and in both summer and winter growth conditions in South Florida (Sutton, Van, and Portier 1992). These data indicate the potential for year-round production of tubers if the monoecious biotype were to become naturalized in water bodies in the southeastern United States. Also, the lack of a strict photoperiod requirement for tuberization in monoecious hydrilla suggests that different management approaches might be required to prevent tuber formation in this hydrilla biotype.

Bensulfuron methyl, currently registered for use in rice, is reported to have great potential to reduce or regulate hydrilla growth and reproduction at relatively low application rates. Anderson and Dechoretz (1988) observed that vegetative growth of monoecious hydrilla was reduced by early postemergent

applications of bensulfuron methyl at 10 ppb or less. Tuber production also was decreased when established hydrilla was exposed to 50 ppb bensulfuron methyl (Anderson 1988). Langeland and Laroche (1990) reported that bensulfuron methyl reduced the growth of hydrilla at very low rates; however, concentrations of 100 ppb or higher were needed to control dioecious hydrilla in Florida.

In the present investigation, we compared responses of monoecious and dioecious hydrilla to various bensulfuron methyl treatments over a period of 6 months. Our main objectives were to determine the duration of the growth-regulating activity of bensulfuron methyl, and possible differential responses between monoecious and dioecious hydrilla to this herbicide in terms of tuber formation.

Materials and Methods

Monoecious and dioecious hydrilla plants were obtained from stock cultures grown over a period of several months in outdoor aquaria. The monoecious hydrilla was established initially from tubers collected from the Potomac River in Virginia. Dioecious hydrilla was established initially from stem apexes from Rodeo Lake in Davie, FL.

The investigation was conducted in 24 outdoor tanks located on the grounds of the Fort Lauderdale Research and Education Center, University of Florida-Fort Lauderdale. The tanks were 0.8 m wide by 2.2 m long (surface area, 1.7×10^{-4} ha) and were filled with pond water to a depth of 0.6 m. Pond water was from the same source as described previously (Van and Steward 1986). Water flow to

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individual tanks was regulated by small petcock valves to provide one water volume change every 24 hr.

The herbicide treatments (concentrations \times biotypes) were arranged as a 4×2 factorial with three replicates and were assigned to the tanks in a complete randomized design.

Hydrilla tubers of both biotypes were allowed to germinate in pond water at 25 °C under continuous light for 3 weeks before planting. Ten sprouted tubers, 10 cm long, were planted in plastic pans 26 cm wide, 30 cm long, and 15 cm deep. The pans were filled with approximately 12 kg of potting soil enriched with 10 g of a slow-release fertilizer (Sierra, 17-6-10 with an 8- to 9-month release). Four pans of a given hydrilla biotype were placed in each tank, and the plants were allowed to grow for 2 weeks prior to herbicide treatment.

At the time of treatment, water flow to individual tanks was stopped, and bensulfuron methyl was applied to the tanks at concentrations of 0, 50, 100, and 200 µg/L. The plants were exposed to the bensulfuron methyl for 4 weeks, after which water exchange was resumed. At 1, 2, 4, and 6 months after herbicide application, one pan of each biotype from each tank was harvested. The harvested biomass was partitioned into shoots, roots, and tubers and oven-dried at 70 °C to a constant weight.

Data for dry weights and tuber numbers were subjected to analysis of variance using a split plot model with herbicide treatments as main plots and harvest dates as subplots. Because of the significant herbicide concentration-plant biotype interactions ($P < 0.05$), the model was reduced, and data for the two biotypes were analyzed separately. Regression analyses of various plant responses over time were then performed for each biotype.

Results and Discussion

Plant dry weight of untreated hydrilla increased according to a third-order polynomial

and reached a maximum weight of 91 and 104 g per pan after 4 months of growth for monoecious (Figure 1a) and dioecious (Figure 2a) hydrilla, respectively. Severe plant damage, including shoot tip reddening and whole plant necrosis, was observed 2 weeks after exposure of hydrilla to all bensulfuron methyl treatments. Growth of both hydrilla biotypes exposed to this herbicide was suppressed for approximately 2 months, resulting in more than 90-percent reduction in plant dry weight in all application rates, as compared to untreated plants at the second harvest in February. These results are consistent with an earlier report by Anderson (1988) of excellent initial herbicidal effect after 1 and 2 months.

Subsequent harvests from the same herbicide treatments, however, revealed various levels of regrowth depending on different treatment rates. For monoecious hydrilla, regrowth was most heavy in the 50-ppb treatment rate, and plants recovered completely after 6 months based on dry weight measurements (Figure 1a). The 100-ppb treatment reduced plant dry weight by approximately 20 percent after 6 months, while sustained low levels of biomass were observed only in the 200-ppb treatment rate over the entire study period.

Bensulfuron methyl also suppressed tuber formation in both monoecious (Figure 1b) and dioecious (Figure 2b) hydrilla at virtually all concentrations tested. The suppression level of tuber formation was often much greater than the corresponding reduction of plant biomass exhibited by the same bensulfuron treatment. For example, tuber production in monoecious hydrilla treated at 100-ppb bensulfuron methyl was reduced by as much as 79 percent after 6 months (Figure 1b), when no more than a 20-percent loss in plant weight was observed (Figure 1a).

The growth-regulating effect of tuber inhibition also appeared to persist long after the plants had recovered from the initial herbicidal effects. The duration of effect of tuber suppression increased with increasing treatment

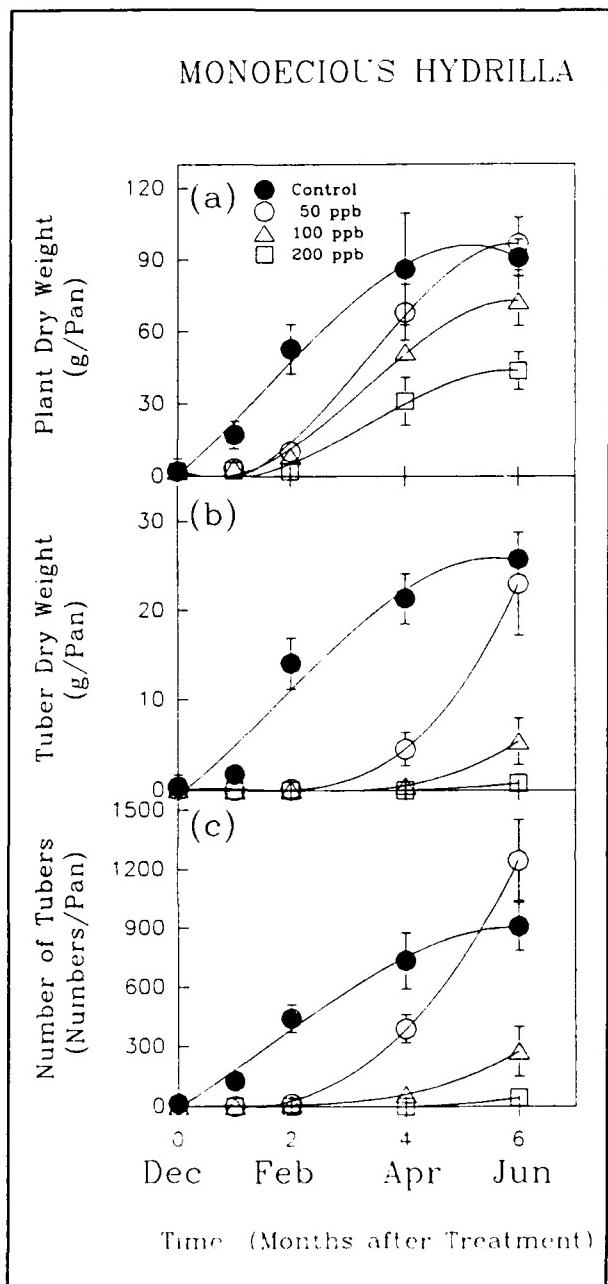


Figure 1. Effects of various bensulfuron methyl treatment rates on vegetative growth and tuber production in monoecious hydrilla. Curves represent best-fitted polynomial regression equations. Vertical lines indicate means ± 1 standard deviation

rates. In monoecious hydrilla, tuber formation was suppressed for a period of approximately 3, 4, and 6 months in the 50-, 100-, and 200-ppb treatments, respectively (Figure 1b). As a result, tuber formation in monoecious hydrilla was reduced by 79 percent in the

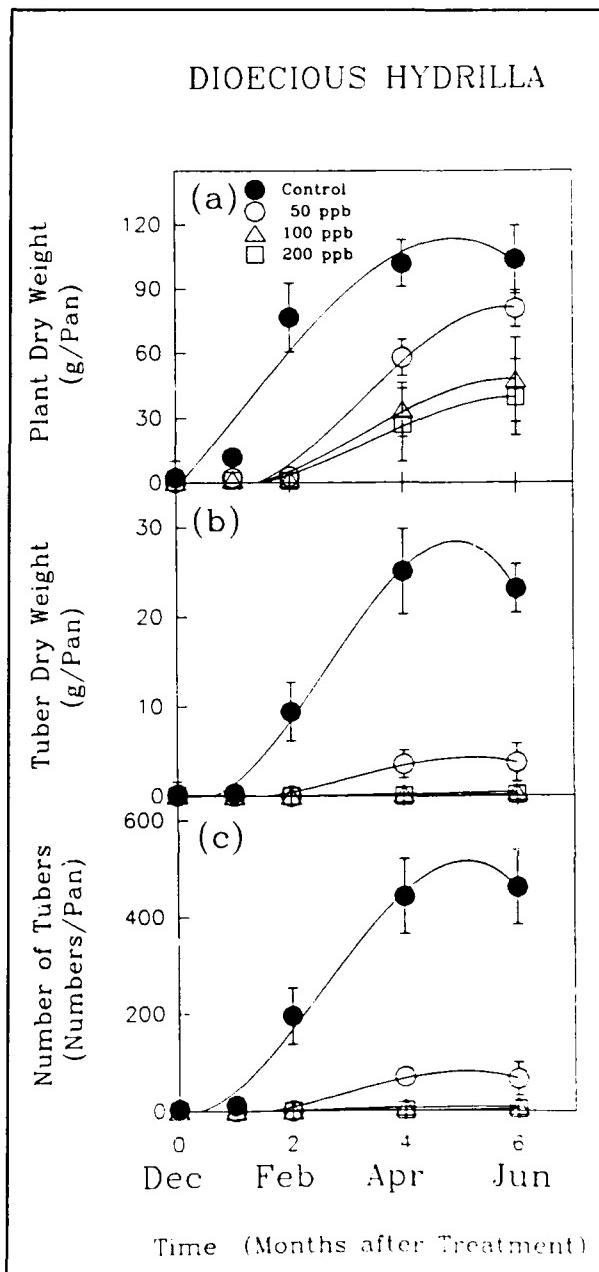


Figure 2. Effects of various bensulfuron methyl treatment rates on vegetative growth and tuber production in dioecious hydrilla. Curves represent best-fitted polynomial regression equations. Vertical lines indicate means ± 1 standard deviation

100-ppb treatment and by 97 percent in the 200-ppb treatment after 6 months. Tuber formation in the 50-ppb treatment was also reduced by 80 percent after 4 months; however, tubers formed rapidly from April to June, so that total tuber weight produced

after 6 months reached values similar to those for tuber weight in untreated plants (Figure 1b).

The ability of monoecious hydrilla to form tubers during the summer months in south Florida was consistent with an earlier report by Sutton, Van, and Portier (1992). An interesting result was the apparent stimulation of tuber formation in monoecious hydrilla treated at 50 ppb after 6 months based on measurements of number of tubers (Figure 1c). The tubers produced in the 50-ppb treatment were much smaller; however, significantly more tubers were formed and, as a result, the total tuber weight in this treatment was similar to tuber weight in untreated plants (Figure 1b). Furthermore, laboratory tests indicated no differences in germinability between these smaller tubers collected from the bensulfuron methyl treatments and those from untreated plants (data not shown).

Untreated dioecious hydrilla began to form tubers 1 month after planting, reaching a maximum number of 444 tubers per pan in April (Figure 2c). Tuber production in dioecious hydrilla apparently ceased from April to June, however, as the tuber curves remained flat during these summer months. This seasonal pattern of tuber production in dioecious hydrilla in Florida was consistent with earlier findings by Haller, Miller, and Garrard (1976) and Sutton, Van, and Portier (1992).

In the 50-ppb bensulfuron methyl treatment, tubers were formed from February to April, but again there was no evidence of tuber production from April to June (Figures 2b and 2c). As a result, tuber formation in dioecious hydrilla was still reduced by 84 percent or more after 6 months in all bensulfuron methyl treatment rates tested. These data suggest that proper timing of bensulfuron methyl applications, based on the seasonal tuber production in dioecious hydrilla, could increase the herbicide performance in this biotype.

Future Work

Based on these results, research planned for fiscal year 1992 will include various studies of timing and multiple applications of low rates of bensulfuron methyl to maintain full-season suppression of plant growth and/or tuber formation in monoecious and dioecious hydrilla biotypes. Responses to bensulfuron methyl treatments of new growth (shortly after emergence from tubers/rootcrowns) in spring applications will be compared to those of established mature vegetation in late summer. Herbicide residues will be monitored in the different bensulfuron methyl treatments, and correlations between chemical residues and corresponding plant responses will be determined.

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Potential Plant Growth Regulators for Aquatic Plant Management

by

Carole A. Lembi¹ and Tara Chand¹

Introduction

For the past several years we have studied the potential of several groups of chemicals to act as plant growth regulators on submersed aquatic plant species. One of the most promising of these groups is the gibberellin synthesis (GS) inhibitors. These compounds inhibit the synthesis of the naturally occurring plant hormone gibberellin. Since gibberellin is responsible for causing plants to increase in length, application of GS inhibitors reduces main stem elongation. The aquatic plant species remain short and nonweedy, and yet are viable and able to provide benefits to the aquatic environment such as oxygen production and habitat.

Using laboratory bioassays (Netherland and Lembi, in press) and outdoor small-scale tests (Lembi and Chand, in press), we have shown that the GS inhibitors flurprimidol, paclobutrazol, and uniconazole are effective (at parts per billion concentrations) in significantly reducing main stem length in hydrilla and Eurasian watermilfoil. Short-term exposures of 2 hr are sufficient to reduce stem length in hydrilla with 75 ppb flurprimidol and in Eurasian watermilfoil with 200 ppb flurprimidol. Stem length remains reduced for at least 28 days following exposure. These data are summarized in Lembi and Netherland (1990) and Nelson et al. (1991).

This report summarizes our results on the dissipation characteristics of flurprimidol in water, sediment, and plant tissues. This information is necessary to determine the environmental fate of GS inhibitors in the aquatic ecosystem, an important consideration in de-

ciding whether these compounds should be further developed for the aquatic market.

Materials and Methods

Metal barrels (67-L capacity) were lined with plastic liners. Loam soil (free from plant growth regulators, herbicides, and other pesticides) was added to a 10-cm depth in each barrel. Approximately 55 L of well water was added, and the suspended soil was allowed to settle. Two healthy milfoil stem apices (10-cm length) without roots were planted in each barrel and allowed to acclimate for 1 week prior to flurprimidol treatment. The milfoil plants were obtained from Martel Pond in Tippecanoe County, Indiana. Flurprimidol (50 percent wettable powder, DowElanco Products Company, Indianapolis, IN) was applied by diluting the compound in approximately 10 ml of water and then stirring the solution into the barrel (without disturbing the soil) to ensure even dispersal. The flurprimidol concentration was 200 ppb active ingredient (a.i.), and the treatment date was June 1, 1990.

Water, sediment, and milfoil samples were taken from each barrel prior to treatment, immediately after treatment, and at 2 hr and 1, 3, 7, 14, and 28 days after treatment. Two barrels were set up for each sampling time; therefore, no barrel was sampled more than once. Water was collected in 1-L polyethylene bottles. Sediment samples were taken using a hand-held, hollow plastic cylinder (5-cm inner diameter) inserted into the sediment to the base of the barrel (10-cm depth). One of the two milfoil plants was collected. The whole plant, including roots, was used for the analysis.

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The procedures for the extraction of flurprimidol from water, sediment, and milfoil tissues and for the detection of flurprimidol using GC and GC/MS are provided in detail in Chand and Lembi (1991).

Results and Discussion

Flurprimidol residue in the water prior to treatment was less than the minimum detectable level of 0.01 ppb. Flurprimidol in water and milfoil tissues showed typical half-life dissipation curves. The half-life in water was 9.1 days and in milfoil, 9.9 days. The compound was detected in the water at 193 ppb immediately after treatment (at 200 ppb) and began to decrease in concentration after that point. Maximum levels of flurprimidol in the milfoil tissue (713 ng/g fresh weight) were detected 1 day after treatment and then began to dissipate. In the sediment, flurprimidol concentrations increased to a maximum of 50 ng/g fresh weight at 7 days and then leveled off with no further dissipation observed over the 28-day period.

At the end of the 28-day sampling period, approximately 89 percent of the flurprimidol had dissipated from the barrel system. Of the 11 percent that remained, the distribution was as follows: 0.02 percent in the plant, 21.3 percent in the upper 5 cm of the sediment column, 0.6 percent in the lower 5 cm of the sediment column, and 78.1 percent in the water. Very little of the compound had apparently leached into the lower portions of the sediment.

We conducted a separate experiment in which barrels were treated with flurprimidol at 1 ppm a.i., and the sediment was sampled over a 6-month period. The flurprimidol concentration in the sediment began to decrease after 28 days. The estimated half-life in sediment was 178 days (almost 6 months).

Also in a separate experiment, flurprimidol, uniconazole, and pacllobutrazol were applied at 1 ppm a.i. The water was sampled over a 56-day period. The half-lives for these com-

pounds in water were 9.3, 5.2, and 24.4 days, respectively.

Our data suggest that flurprimidol dissipates rapidly from the water, probably through photodegradation, since this compound is susceptible to this breakdown mechanism (Lilly Research Laboratories 1983). The compound is rapidly taken up by the plant. Although the dissipation curve in the plant tissue is similar to that of water, the half-life was slightly longer than that in water. The difference between 9.1 and 9.9 days may not be significant, but this suggests that the compound in the plant may not be totally exchangeable with the water. Apparently, only a very small tissue concentration (perhaps as low as 3 to 5 ng/g fresh weight) is required to achieve main stem reduction (data not presented). At an initial internal concentration of 713 ng/g fresh weight, this threshold level would not be achieved until 71 to 78 days after treatment (without water exchange after treatment). Also, the plant may take up flurprimidol from the sediment where it can remain for a long period of time, although the potential for sediment uptake remains to be tested.

Rapid dissipation from the water, retention in the sediment, and the potential for either plant uptake from the sediment or long-term retention in the plant tissue would be ideal conditions for long-term main stem reduction of hydrilla and Eurasian watermilfoil with flurprimidol.

Future Work

The results from these dissipation studies suggest additional research that is necessary to understand the fate of flurprimidol in its unaltered state and to determine the potential for long-term stem reduction. First, we need to know the potential for breakdown in water under low light intensities where photodegradative processes are minimal. Will the compound be susceptible to nonphotodegradative processes such as hydrolysis or microbial breakdown in the deeper portions of the water column? As indicated above, we also

need to know whether the plant can take up flurprimidol from the sediment or whether uptake is strictly from the water followed by long-term retention in the plant tissue. This can be accomplished using ¹⁴C-flurprimidol, which has already been supplied to us by DowElanco.

Eventually, the GS inhibitors should be tested in the field, hopefully at the Lewisville facility. Part of these studies should be devoted to confirming effective exposure times and dosages suggested by the laboratory and barrel experiments. The potential for long-term stem reduction (longer than 28 days) at low exposure times and concentrations must be tested, as should the potential for tuber formation in treated hydrilla. Additional long-term goals with the GS inhibitors are to investigate the susceptibility of nontarget plant species to these compounds, the habitat value of height-reduced plant stands, and toxicology to nontarget fauna.

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Phenology of Aquatic Plants

by
John D. Madsen¹

Introduction

Phenology is the study of seasonal changes in the morphology, physiology, and resource allocation of organisms. Applied to target nuisance aquatic plants, this includes the analysis of growth rates, physiological processes, and allocation of biomass, carbohydrates, and nutrients. Although the acquisition of this information alone is worthwhile, our goal is to directly apply this knowledge to the control of target aquatic plants by finding weak points in their seasonal growth cycle for the application of control methods, either singly or in concert. With this information to assist in management planning, management of these species may be more effective, less costly, and have reduced environmental impacts.

In the past year, research has focused on the phenology of waterhyacinth (*Eichhornia crassipes*). Two experiments were performed to evaluate seasonal growth rates, biomass development, and allocation patterns in this nuisance floating species.

Methods and Materials

Experiments in 1991 were conducted using two ponds at the Lewisville Aquatic Ecosystem Research Facility in Lewisville, TX. Both ponds were amended with hydrochloric acid (to maintain a pH below 8.0) and Aquashade at 1 ppm (to reduce algal growth). One pond was further amended with 11.4 kg of ammonium sulfate fertilizer per week (referred to as the nitrogen pond), while the other was not amended with fertilizer (reference pond).

Ring study

The ring study examined the development of waterhyacinth at a given location across time. The rings were 1 m² in size, made of wire mesh with floats to provide buoyancy and attached to wire supports to maintain their position in the middle of the pond at a depth of approximately 0.5 to 1 m. The ring study was initiated by "planting" five small rosettes of waterhyacinth per ring. Three cohorts or time periods were used: cohort A began 27 May, cohort B began 23 July, and cohort C began 24 September.

Three rings were harvested for each sampling period for each cohort; cohorts were sampled at 0, 1, 2, 3, 5, 7, 9, 12, 15, 18, 22, and 26 weeks after the start of the cohort. Data presented here will be through mid-October.

Plants were harvested, the number of mature plants counted, and the plants sorted to component plant parts, dried, and weighed to determine biomass.

Run study

The run study was designed to examine the spatial development of waterhyacinth mats. Eighteen runs were used in each of the two ponds, with each run being 1 m wide and 4 m long. All eighteen runs in both ponds were planted on 29 May. Samples were taken 5, 7, 9, 11, 13, and 15 weeks after planting, at which time three randomly selected runs were sampled. Samples were taken at the back edge, the front edge, and in the middle of the mat using a 0.1-m² quadrat, with plants

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counted, separated into shoots and roots, dried, and weighed to determine biomass.

Results and Discussion

Ring study

Waterhyacinth increase in density followed a typical sigmoid-growth curve, with the nitrogen pond populations showing the most rapid increase in cohort A and the reference population in cohort B (Figure 1). No significant increase in cohort C was observed because of the short time period since initiation (similar to the results observed for both populations in both cohorts A and B after 3 weeks of growth).

Each population exhibited a 3-week lag phase in growth ("colonization"), a rapid phase of increase in density, and then a saturation phase when density leveled off or decreased.

This saturation level varied from 175 rosettes m^{-2} in the nitrogen pond to 200 rosettes m^{-2} in the reference pond, the variation due to the difference in average size of the rosettes.

Changes in density of adult plants can partially be explained by changes in the density of daughter plants, those vegetative propagules produced by mature plants (Figure 2). Initially, daughter plant densities remain low during the lag phase of 3 weeks. Then, daughter plant densities increase rapidly, but begin to decline as the population approaches mature plant stable densities.

Although some of the density stability observed in mature stands of waterhyacinth was due to mortality of mature plants, most of this stability appeared to be density-dependent control of daughter plant production. Further insight into this mechanism may have potential for management. For instance, some

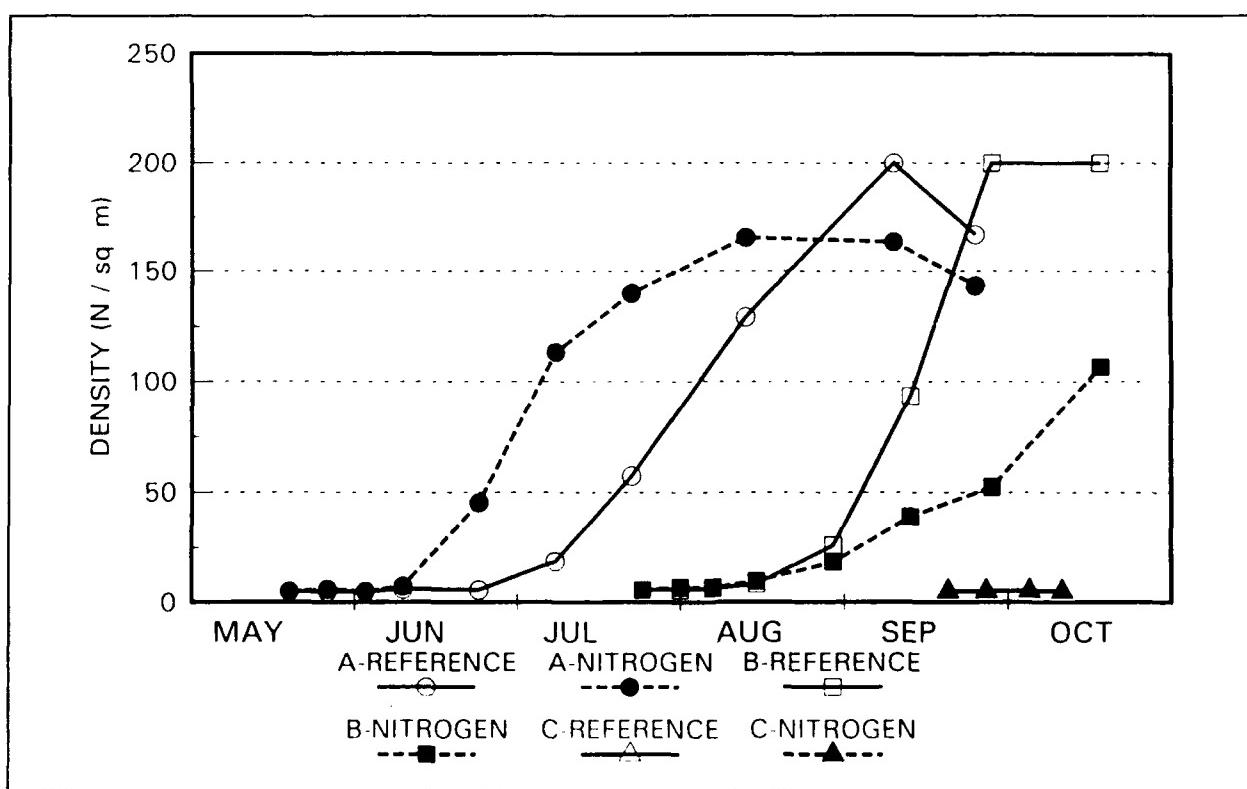


Figure 1. Mature rosette density (rosettes m^{-2}) of three cohorts of waterhyacinth grown in 1- m^2 rings in reference and nitrogen ponds during 1991 growing season

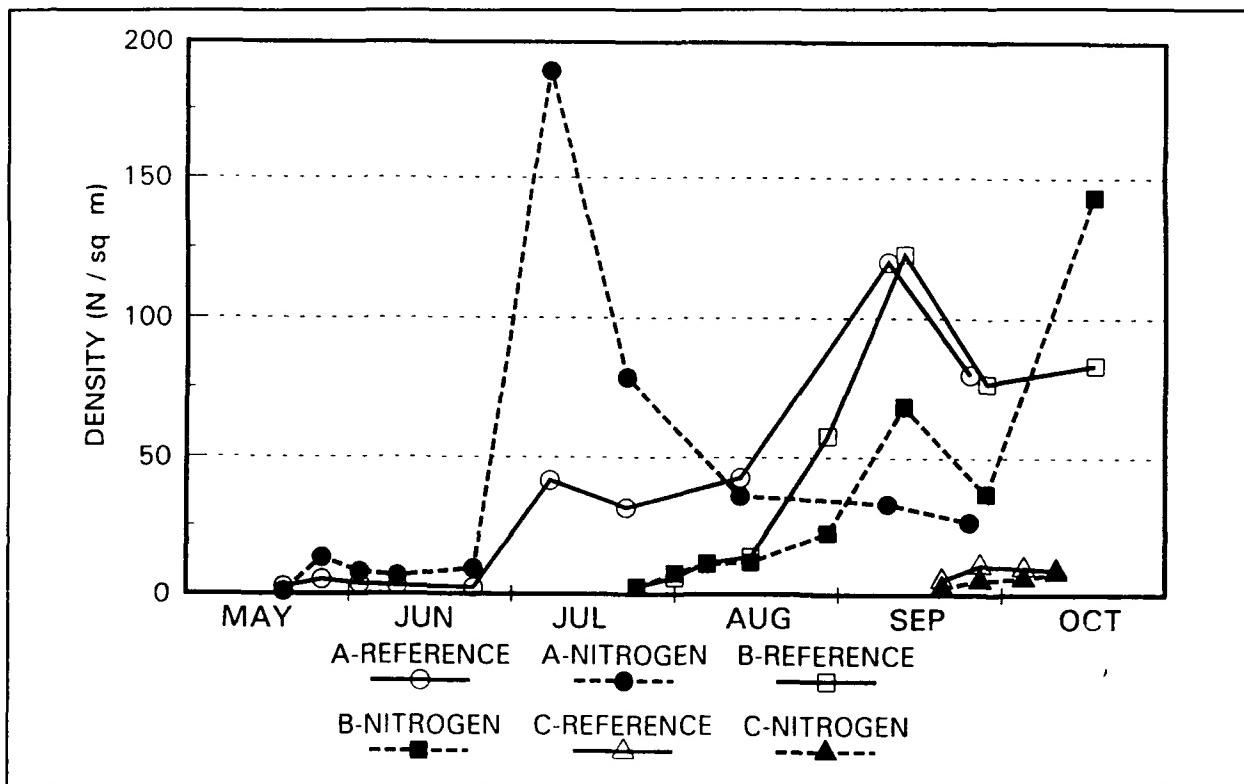


Figure 2. Daughter plant density (rosettes m^{-2}) of three cohorts of waterhyacinth grown in 1-m^2 rings in reference and nitrogen ponds during 1991 growing season

plant growth regulators could inhibit daughter plant production and thus effectively prevent a population from entering the growth phase of the population cycle.

Biomass levels follow a pattern similar to that observed for density (Figure 3). Weight of individual plants remains low early in the growth cycle, then increases rapidly during the growth and stationary population phases, resulting in continued increases in total biomass. Peak biomass observed for waterhyacinth was $2,000 \text{ g m}^{-2}$ dry weight.

Biomass allocation patterns are an important, yet neglected, aspect of the growth of new populations. One particularly evident allocation pattern in waterhyacinth is the percent of biomass allocated to root tissue (Figure 4). Root tissues are particularly important for waterhyacinth in acquiring nutrients from the water column or hydrosoil.

Early in the growth phase, most of the new biomass was allocated to the production of roots. As much as 70 percent of plant biomass was composed of root tissue. As density began to increase and the plant entered the growth phase, proportionately more material was allocated to shoot material. The percentage of total biomass going to roots continued to decrease in the stable phase of the growth cycle, although total root biomass continued to increase as long as total biomass increased. In mature stands, as little as 30 percent of total biomass was composed of roots, although this figure is still substantially higher than is typical of most aquatic or terrestrial herbaceous plants.

Run study

The results from the run study were consistent with those of the ring study, so additional graphs will not be presented on the same

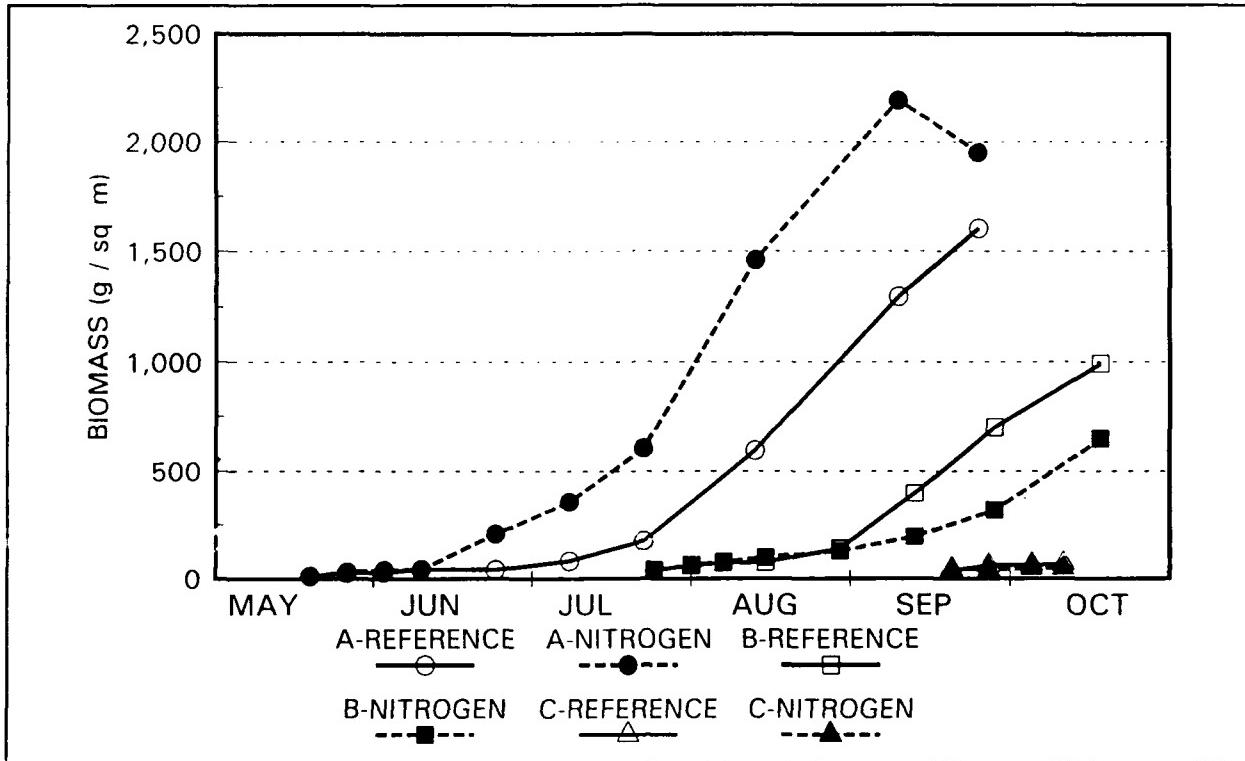


Figure 3. Total plant biomass ($\text{g dry weight } \text{m}^{-2}$) of three cohorts of waterhyacinth grown in $1-\text{m}^2$ rings in reference and nitrogen ponds during 1991 growing season

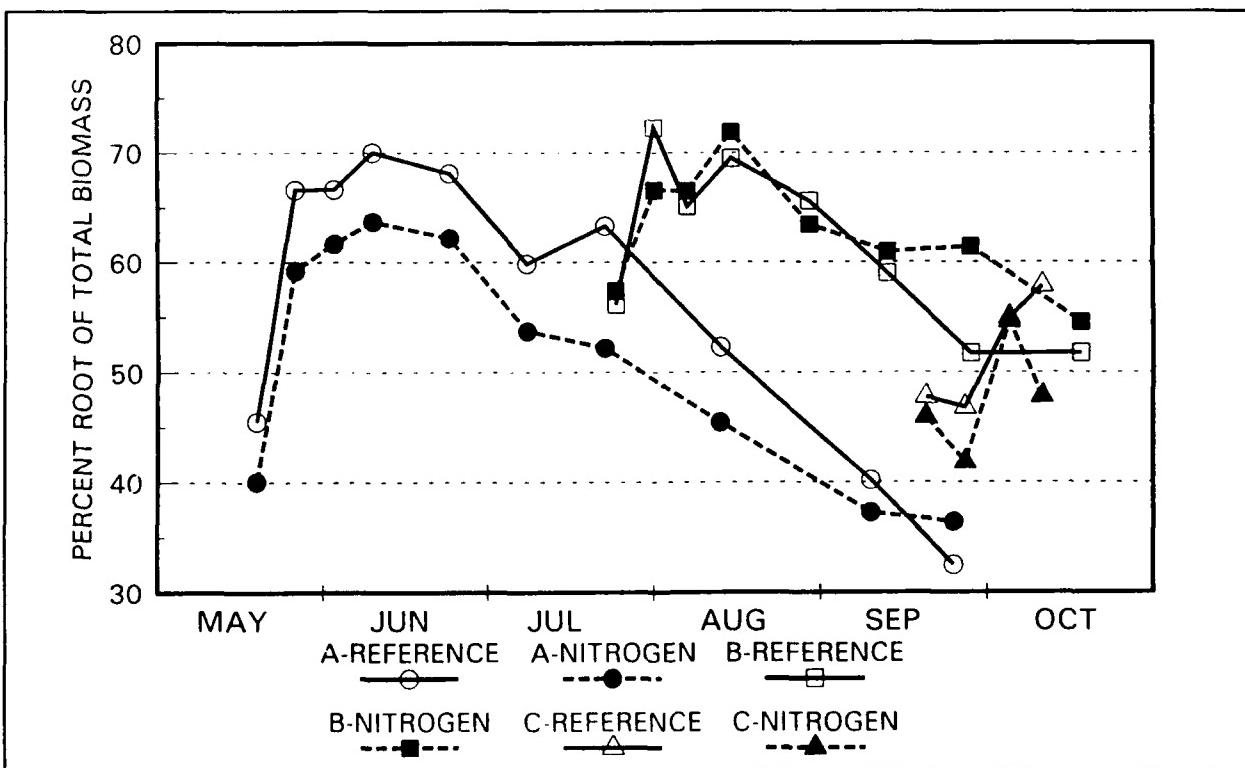


Figure 4. Percent allocation of total biomass to roots for three cohorts of waterhyacinth grown in $1-\text{m}^2$ rings in reference and nitrogen ponds during 1991 growing season

parameters. The consistency of the results indicated that temporal and spatial changes in developing mats are similar. Many of these changes can be attributed to density-dependent changes in the morphology and growth of plants, similar to the 3/2 thinning rule.¹

In Figure 5, the density of rosettes is plotted against the average weight per shoot of biomass samples from the run study. Colonization of a site begins with small plants at low density. These plants increase in numbers and density without increasing in size, until they produce a new mat, indicated by the row of edge samples along the x-axis. These plants slowly increase in size and density, after the density has reached approximately 150 m^{-2} . As a mature dense mat is formed, individual size continues to increase, but density decreases as the result of intraspecific competition.

The significance of the dotted line in Figure 5 is as a theoretical boundary of average size versus density across which the population cannot pass. Once this boundary is reached, individual size must increase for plants to be successful against their neighbors, and mortality of the smaller plants results in reduced density.

The stages in mat development both temporally and spatially are indicated in Table 1. The table separates the phases of growth as invasion, edge, middle or transition, and mature; however, these phases are somewhat arbitrary since these are in reality points or segments along a continuum of change.

During invasion, plants are typically small daughter plants. Density is low, and plants are small, with short bladder leaves for flotation. Root allocation is equal to shoot allocation,

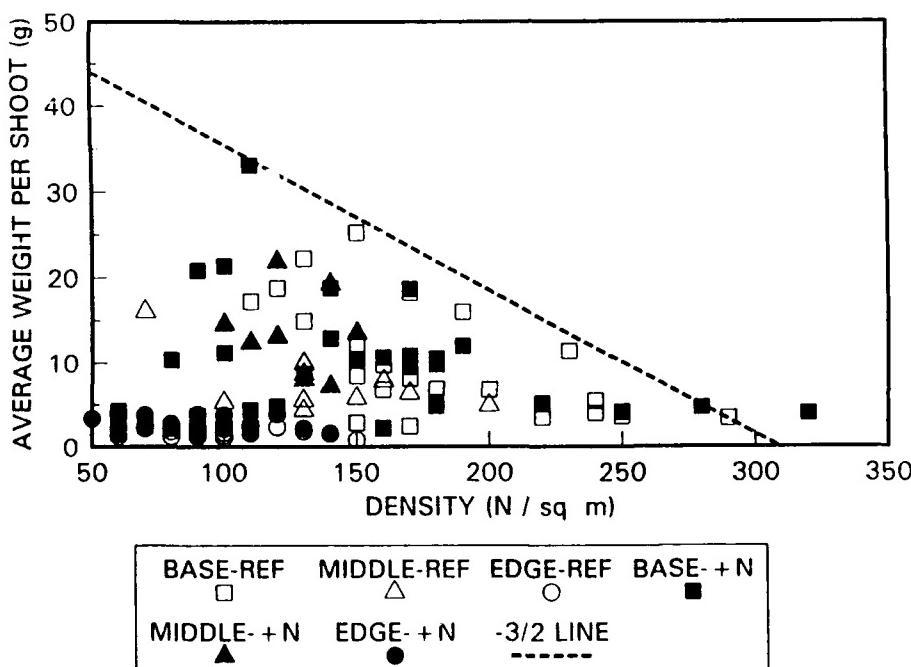


Figure 5. Relationship between density (rosettes m^{-2}) and average plant weight (g dry weight) for samples from the base, middle, and edge of mats grown in runs from both reference and nitrogen ponds

¹ A. R. Watkinson. 1986. Plant population dynamics. In *Plant ecology*. M. J. Crawley, ed. 137-184. Oxford, England: Blackwell Scientific Publications.>

Table 1
Waterhyacinth Mat Development—Temporally and Spatially

Characteristic	Invasion	Edge	Middle	Mature
Density	Low	Increasing	Peak	Decreasing
Plant size	Small	Small	Slow increase	Rapid increase
Root allocation	Equal	Increasing	Stable	Declining
Leaves	Floating	Floating	Transition	Erect
Biomass	Low (200-400 g)	Low (200-600 g)	Moderate (1-2 kg)	High (2-4 kg)
Propagation	Vegetative	Vegetative	Both	Flowering

and propagation is largely vegetative. Edge plants are similar to those in the invasion phase, except there is an increase in density and a substantial increase in root allocation.

During the middle phase of growth, density peaks and there is a slow increase in average plant size as a result of intraspecific competition and the initiation of self-thinning effects. Root allocation stabilizes, then begins to decline. Leaves begin the transition to the erect form, as the mass of the mat alone provides the buoyancy needed to remain on the surface. Although flowering becomes more evident, vegetative propagation still occurs, and these plants are important to the support of daughter plants farther out in the mat.

In a mature stand, density decreases from the effects of self-thinning. Intense competition results in the rapid increase of average plant size, with only erect leaves found on these plants. Root allocation continues to decline in mature stands. Flowering becomes the important mode of propagation, since daughter plants are not produced, with the exception that the stem bases of mature plants are the most important for overwintering.

A better understanding of waterhyacinth mat dynamics will result in better management planning. Early invasion-stage plants are smaller, and thus have lower reserves of carbohydrates. Targeting early invasion stages would result in higher control efficien-

cies and reduced treatment costs. Early applications of biocontrol agents would also improve their effectiveness, since there is less biomass to control, and growth rates at the early stages are slower.

Future Research

Waterhyacinth

Future research on waterhyacinth will include additional studies of mat dynamics and the effects of different starting colony sizes on growth rates. In addition, planning for a small-scale trial of a phenological management technique is scheduled during calendar 1992.

Eurasian watermilfoil

Eurasian watermilfoil studies will be initiated in calendar year 1992, including studies of seasonal biomass, growth, and allocation of carbohydrates and resources. In addition, studies of the seasonality of spread by runners and fragments is being planned, following the design of a similar study in a northern temperate lake.¹ Separate experiments are also being planned to examine the early invasion and establishment process, and to evaluate the importance of spread by seed. The latter is especially significant in that no current experimental information is available; however, an increasing body of observation indicates that this process might be a significant mode for remote areas.

¹ J. D. Madsen, L. W. Eichler, and C. W. Boylen. 1988. Vegetative spread of Eurasian watermilfoil in Lake George, New York. *Journal of Aquatic Plant Management* 26:47-50.

Seed Germination and Seedling Survival of Waterhyacinth

by

Rebecca S. Westover¹ and John D. Madsen¹

Introduction

Waterhyacinth (*Eichhornia crassipes*) is a perennial mat-forming rosette species originally from tropical and subtropical South America. It is an invasive nuisance species in the southern United States, where it forms a dense mat that prevents human use of waterways and competitively excludes native submersed and floating-leaved plants.

In most localities, vegetative propagation and overwintering is the predominant mode of both spread and regrowth from one year to the next. However, the plants are capable of producing large numbers of seeds, and seedlings have been observed at a few sites in nature.² Seeds will readily germinate in the lab and under controlled field conditions.

Sexual propagation of waterhyacinth has received little research attention; however, anecdotal information indicates that regrowth from seeds may explain why the plant returns after apparently successful control procedures. An understanding of the complete life cycle of this plant is needed if we are to more successfully control its growth.

Methods

Pond experiment

Pond 19 held a waterhyacinth seed bank in the sediment from phenology research conducted in 1989. The pond was flooded early in 1990 and contained water throughout the summer and fall of 1990. Seedlings germinated underwater to depths up to 1 m, then

abscised from the tap root after 2 to 4 weeks of development, causing seedlings to float. Seedlings were counted throughout the growing season. Seedlings were not marked, so data on specific recruitment and mortality were not available.

Pond 42 has been used as a culture pond for waterhyacinth since 1989. A large mat was formed across the back of the pond in 1989 and 1990. Water levels caused test seedlings to grow in the mud flats at the periphery of the pond. Seedlings were counted and marked as they emerged. In 1991, organic matter was removed from test plots. All seedlings from both ponds were monitored throughout the growing season.

Greenhouse experiment

Sediment from greenhouse-grown plants was placed in growth flats (11 by 21 by 2 in.). The sediment was treated as follows:

- a. Dried for 8 weeks.
- b. "Planted" fresh in early spring.
- c. Stored wet for 28 weeks in cold, but not freezing, conditions.
- d. Stored dry for 28 weeks in cold, but not freezing, conditions.
- e. "Planted" fresh in late spring.

All seedlings were marked as they emerged and monitored throughout the growing season. The growing flats were kept wet during the experiment.

¹ US Army Engineer Waterways Experiment Station, Lewisville Aquatic Ecosystem Research Facility, Lewisville, TX.

² S. C. H. Barrett. 1980. Sexual reproduction in *Eichhornia crassipes* (water hyacinth). II: Seed production in natural populations. *Journal of Applied Ecology* 17:113-124.

Results and Discussion

Pond experiment

Seedling counts were much higher in test areas where organic material was removed (1991) than in undisturbed areas (Figure 1). Also, the contributions of additional seeds to the seed bank may have resulted in higher seedling emergence in 1991 than 1990. Seedling enclosure areas from 1990 did not produce seedlings in 1991.

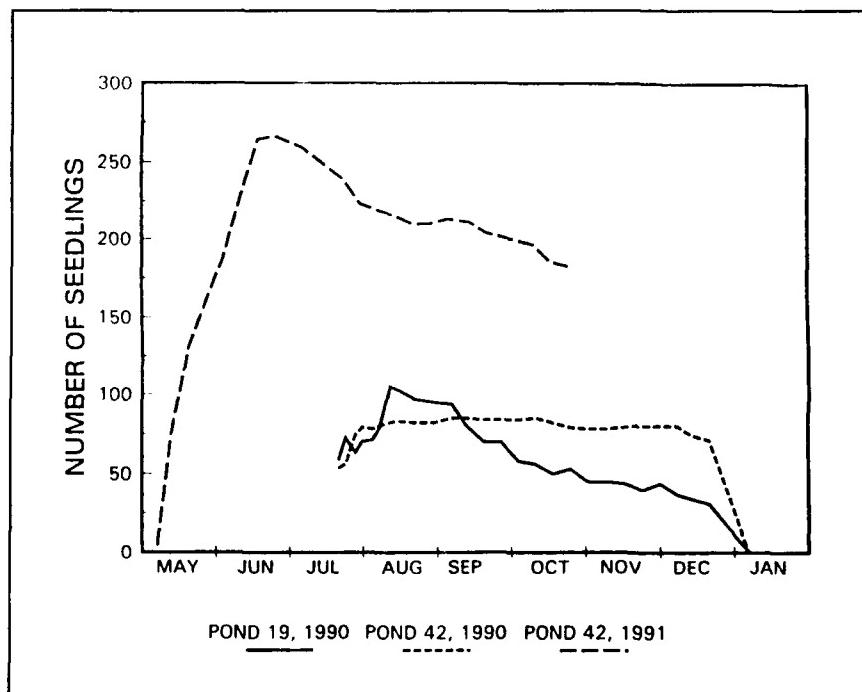


Figure 1. Total numbers of seedlings observed in Pond 19 and 42 enclosures during 1990, and in Pond 42 enclosures during 1991

Recruitment occurs predominantly in the spring. Only minor additional germination occurred in midsummer or early fall (Figure 2). Mortality of seedlings was lower when water levels remained constant (Figure 2). Stable water levels contribute to higher seedling survival rates. Mortality was sporadic, with increases occurring during times of rapid water loss, as a result of seedlings being caught on surrounding vegetation with roots exposed to the air. However, mortality was primarily

caused by temperature drop at the first major freeze of the winter (Figure 2).

Seedlings did not flower until mid-September (Figure 3), which was a 4-month lag behind vegetatively reproducing plants that flowered in mid-May (unpublished data).

Greenhouse experiment

Dried seeds had lower viability than seeds remaining wet (Figure 4). Sediments remaining wet from early and late spring had comparable seedling emergence frequencies; sediments stored wet were intermediate to sediments stored dry. Recruitment occurred predominantly in the spring. As with the pond experiment, seedling emergence occurred early in the growing season, but greenhouse emergence was not as synchronized as pond emergence, possibly because of lower light levels.

Conclusions

Significant numbers of waterhyacinth seedlings were observed in two of the study ponds in 1990. Subsequent study has indicated several aspects of seed germination and seed-

ling survival with important implications for management of this species.

- Recruitment, survival, and development were superior on shallow mudflat environments compared to submersed pond bottom environments.
- The removal of dead organic material from the mat increased the number of seedlings in the area of the disturbance.

- During the growing season, water-level fluctuations increased seedling mortality.
- The time span from seedling emergence to flowering was approximately 100 to 120 days.
- Freezing waterhyacinth seeds did not inhibit germination; however, drying of the seeds did reduce germination rates. Storage of waterhyacinth seeds also resulted in reduced germination rates.

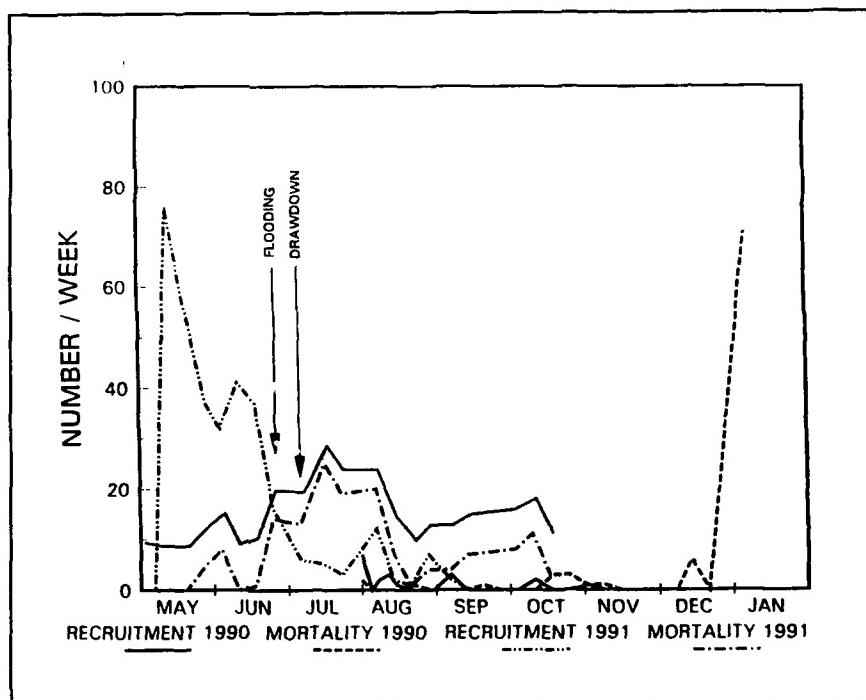


Figure 2. Waterhyacinth seedling recruitment and mortality in Pond 42 enclosures during 1990 and 1991

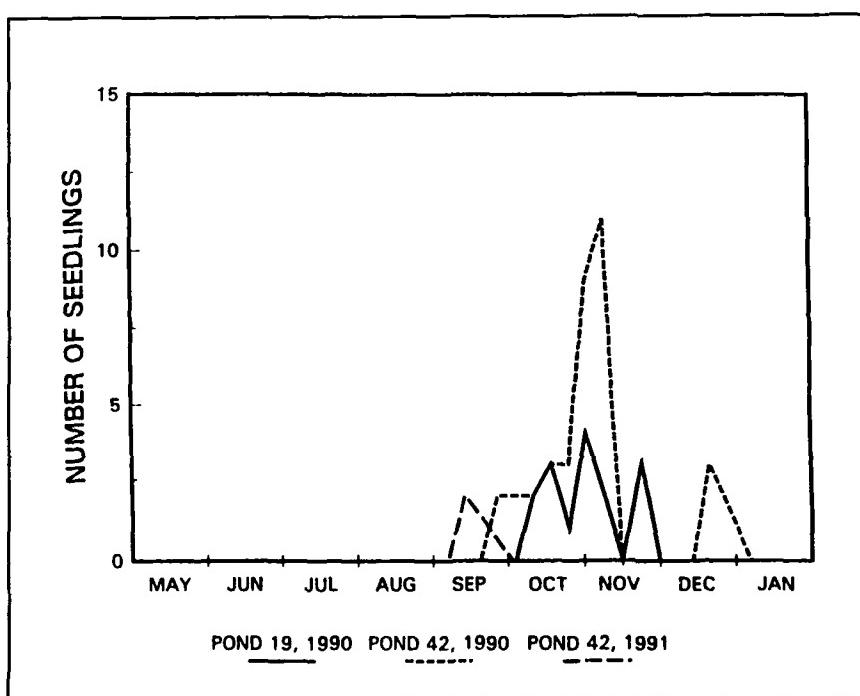
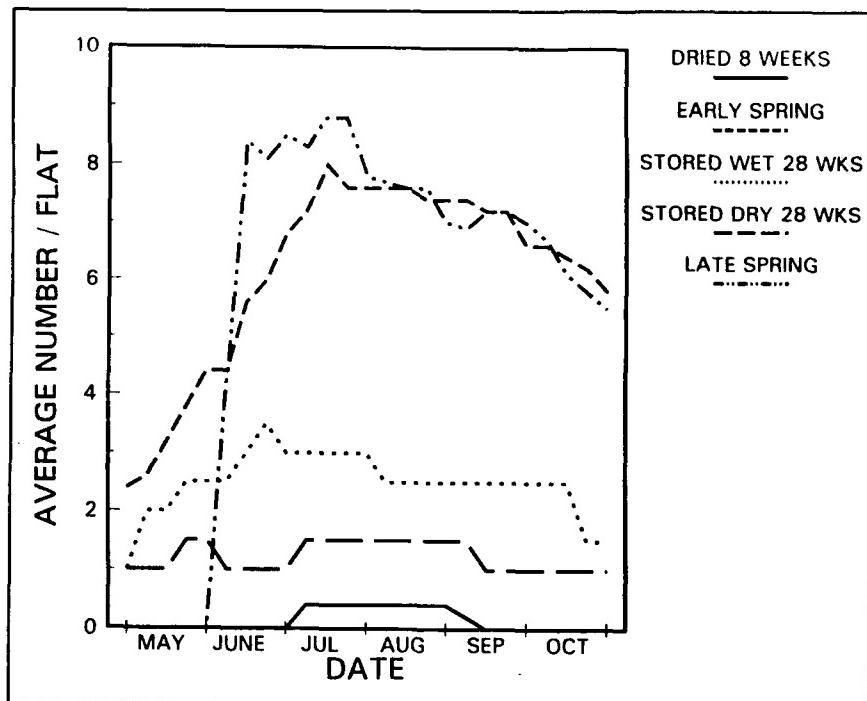


Figure 3. Flowering of waterhyacinth seedlings in Ponds 19 and 42 during 1990 and in Pond 42 during 1991

Although a strictly annual life cycle of waterhyacinth could not produce nuisance problems within a given year, sexual propagation may allow populations on the fringe of its range to overwinter particularly bad years. In addition, the seed bank of waterhyacinth may allow delayed regeneration of waterhyacinth within the same growing season as an otherwise effective control application, as well as in subsequent years. If a large and potentially successful seed bank is present, a management strategy that incorporates



*Figure 4. Average numbers of seedlings in greenhouse flats
for five treatments*

pre- or post-emergent control of waterhyacinth seedlings may prolong the efficacy of the management treatment.

Acknowledgment

Keith Loyd provided assistance in the field and greenhouse.

Ecology of Aquatic Plants

Overview of Ecological Investigations: The Role of Sediment Composition

by
John W. Barko¹

Introduction

Within the Aquatic Plant Control Research Program (APCRP), we have for a number of years studied the ecology of submersed aquatic macrophytes. These studies have focused not only on environmental factors affecting the growth and distribution of aquatic macrophytes, but also on ways in which macrophytes influence their environment. Interactions among key limiting factors (light, inorganic carbon, and nutrients) have been examined in detail, both in a laboratory setting under controlled environmental conditions and in the field.

Among the multitude of nutrients required by submersed aquatic macrophytes, nitrogen (N) appears to be most critical (see below). Nitrogen is derived primarily from the sediment, rather than from the water column. Thus, the availability of sediment N, particularly under conditions of adequate light and inorganic carbon (for photosynthesis), is very important in influencing submersed macrophyte growth. Under many circumstances, uptake by these macrophytes can significantly deplete sediment N pools. Subsequent increase in sediment N availability appears to depend primarily upon mineralization of organic matter in sediment and inputs of N through sedimentation.

During approximately the past 5 years, ecological research within the APCRP has been expanded to consider complex interactions among environmental factors and submersed macrophyte growth. Most recent studies have focused on mechanisms whereby submersed macrophyte communities influ-

ence their environment. It is now apparent, based on results of recent field and laboratory investigations (see below), that submersed macrophytes play a very active role in affecting environmental conditions, particularly within the sediment, in ways that potentially determine the duration and extent of nuisance growth conditions.

The objectives of this article are to provide an overview of macrophyte nutritional ecology, summarize effects of sediment composition on macrophyte growth, and consider related management implications and associated recommendations.

Macrophyte Nutritional Ecology

For many years, considerable controversy has persisted regarding the role of roots versus shoots and sediment versus open water in the nutrition of submersed aquatic macrophytes (reviewed by Sculthorpe 1967; Denny 1980; Smart and Barko 1985; Agami and Waisel 1986; Barko, Adams, and Clesceri 1986). Phosphorus and nitrogen have been studied most extensively, and for these nutrients, sediment is the primary source for uptake. Sediment appears to be the principal site for uptake of iron, manganese, and micronutrients as well. These elements tend to coprecipitate and are usually present in extremely low concentrations in oxygenated surface waters.

Dissolution products of relatively abundant salts are taken up principally from the open water. Among these ions, potassium and calcium are potentially most important in affecting submersed macrophyte growth. Potassium can be obtained from the sediment,

¹ US Army Engineer Waterways Experiment Station, Vicksburg, MS.

but is taken up by submersed macrophytes most abundantly from the open water (Barko 1982, Huebert and Gorham 1983, Barko et al. 1988). Under some conditions this element may be exchanged by submersed macrophyte roots for ammonium ions in sediment (Barko et al. 1988). Calcium is a component of the carbonate system and plays an important role in photosynthetic bicarbonate utilization (Lowenhaupt 1956, Smart and Barko 1986).

Emphasis in this paper is placed on phosphorus (P) and nitrogen because these elements have the greatest potential for influencing macrophyte production in aquatic systems.

A simple empirical model for predicting the relative contribution of sediments and open water to the P economy of submersed macrophytes has been developed by Carignan (1982). In application, the model predicts >50 percent of the supply of P to submersed aquatic macrophytes from sediments where the ratio of dissolved reactive phosphorus (DRP) in the sediment interstitial water to DRP in the open water exceeds about 4. Concentrations of soluble P in the surface waters of lacustrine systems supporting submersed aquatic macrophytes rarely exceed 10 µg/L (Patterson and Brown 1979). Indeed, concentrations approaching about 20 µg/L can cause the exclusion of submersed macrophytes due to light attenuation associated with stimulated algal growth (Jupp and Spence 1977; Phillips, Eminson, and Moss 1978; Sand-Jensen and Sondergaard 1981). From Carignan's model, it is apparent that at 10 µg/L DRP in the open water, as little as 40 µg/L DRP in sediment would provide approximately half the P supplied to submersed aquatic macrophytes.

In evaluating submersed macrophyte growth in relation to specific properties of compositionally diverse sediments, Barko and Smart (1986) reported DRP concentrations in the sediment interstitial water ranging from 40 to >9,000 µg/L, with an overall average of 1,150 µg/L. These results, in combination with predictions of Carignan's model, suggest that P acquired from most sediments probably accounts for much greater than 50 percent of

total P uptake by rooted submersed macrophytes. Results of in situ investigations conducted in a mesotrophic region of Lake Memphremagog indicated that the contribution of P from sediments to rooted submersed macrophytes can approach 100 percent (Carignan and Kalff 1980). Chambers and Prepas (1989) reported that submersed macrophytes growing in rivers, even in infertile, coarse-textured sediments, obtained >70 percent of required P from the sediment. In laboratory studies, production of submersed macrophyte biomass in excess of 1 kg dry mass m⁻² has been achieved routinely on sediments with no P in solution (Smart and Barko 1985).

Owing to the large exchangeable pool of P in most lake sediments (Carignan and Flett 1981), it is unlikely that submersed macrophytes are often limited in their growth by P availability. Indeed, attempts to stimulate submersed macrophyte growth in situ by P addition to sediment (e.g., Anderson and Kalff 1986, Moeller and Wetzel 1988) or to retard growth by reducing sediment P availability (Mesner and Narf 1987) have been unsuccessful. This generalization, however, may not apply to extremely infertile sediments. For example, the growth of *Littorella uniflora* on sand in oligotrophic Lake Hampen, Denmark, was limited by the availability of sediment P (Christiansen, Skovmand Friis, and Sondergaard 1985).

Great attention to the P economy of submersed aquatic macrophytes reflects the unparalleled importance of this nutrient in the eutrophication of lacustrine systems (Schindler 1974, 1977). Given the demonstrated capacity of these macrophytes to take up P directly from sediments, vegetation of the littoral zone needs to be viewed as a potential source of this nutrient to other components of the aquatic environment (Barko and Smart 1980, Carignan and Kalff 1980, Smith and Adams 1986, Moeller and Wetzel 1988).

Information on the relative contributions of sediment and open water to the N economy of submersed macrophytes is quite limited in comparison with that available for P. However, a

few experiments incorporating use of the ^{15}N isotope have demonstrated that this element can be supplied to submersed macrophytes readily from both the sediment and the open water (Nichols and Keeney 1976a, Short and McRoy 1984). These experiments indicated collectively that uptake rates were proportional to N concentration in respective sediment or open-water mediums, and that the studied macrophyte species preferred ammonium over nitrate as the form of N. Since the concentration of ammonium-N in sediment is usually much greater than in the open water of lacustrine systems (Nichols and Keeney 1976b), sediment appears to provide the major source for N uptake by rooted submersed macrophytes. However, this generalization may not apply to enriched riverine systems, such as the Potomac River, where ammonium-N concentrations routinely exceed 100 $\mu\text{g/L}$ in macrophyte beds (Carter et al. 1987).

In any event, it is clear, based on laboratory studies, that submersed macrophytes can satisfy their N requirements over at least the short term by uptake exclusively from sediments (Huebert and Gorham 1983, Barko and Smart 1986).

In contrast to results of in situ P fertilization experiments, yielding little or no effects (see above), fertilization of sediment by the addition of N alone or in combination with other elements has significantly increased the growth of submersed macrophytes both in freshwater (Anderson and Kalff 1986, Duarte and Kalff 1988, Moeller and Wetzel 1988) and marine systems (Orth 1977, Bulthuis and Woelkerling 1981). These results suggest that the availability of N in sediments may under some circumstances limit the growth of submersed macrophytes.

Based on studies of ammonium availability and eelgrass growth, Dennison, Aller, and Alberte (1987) suggest that concentrations of ammonium-N at levels less than about 140 $\mu\text{g/L}$ in the sediment interstitial water may result in N-limited growth. Pools of ammonium-N in sediment interstitial water appear to be buffered by smaller exchange-

able pools than for P (Carignan 1985, Barko et al. 1988, Chen and Barko 1988). Thus, N is depleted from sediments much more rapidly than P, and is more likely than P to limit production of submersed macrophytes.

Effects of Sediment Fertility on Macrophyte Growth

In extensive laboratory investigations, Barko and Smart (1986) demonstrated relatively poor growth of *Hydrilla verticillata* and *Myriophyllum spicatum* on highly organic sediments and sands compared with growth on fine-textured inorganic sediments. The growth of these species decreased almost linearly with increasing sediment organic matter up to a concentration of about 20 percent. From fertilization experiments, these researchers concluded that macrophyte growth limitation on sands and organic sediments resulted from nutrient deficiencies.

Since organic matter and sand (i.e., coarse-textured sediment) have opposing influences on sediment density, their effects on macrophyte growth can be generalized as a function of sediment density. Sands possess high bulk density and low nutrient availability. However, the actual fertility of sands may vary considerably in nature with groundwater nutrient inputs to the root zone (cf. Fortner and White 1988, Lodge et al. 1988). Organic sediments possess low bulk density, and their nutrient content (commonly considered to be high) is actually quite low on the basis of sediment volume (DeLaune, Buresh, and Patrick 1979; Barko and Smart 1986).

Nutrient uptake by rooted submersed macrophytes growing on low-density organic sediments is potentially hindered by the long distances over which nutrients must diffuse (cf. Barko and Smart 1986). In addition, nutrient availability in these sediments can be limited by complexation with organic matter (Sikora and Keeney 1983). Macrophyte nutrition on highly organic sediments may also be disrupted by the presence of phytotoxic compounds produced during anaerobic decomposition (Drew and Lynch 1980).

Additions of labile organic matter to sediments at low levels, for example from the precipitation of algal detritus (Moeller and Wetzel 1988), may provide nutritional benefits to submersed macrophytes, particularly on coarse-textured sediments in oligotrophic systems (Sand-Jensen and Sondergaard 1979, Kiorboe 1980). However, the accumulation in sediments of large amounts of refractory organic matter, with potential inhibitory properties (Barko and Smart 1983), can generally be expected to diminish sediment nutrient availability and associated growth of rooted submersed macrophytes.

During the aging of lakes, an increasing proportion of sediment organic matter derives from emergent vegetation (Godshalk and Wetzel 1978, Godshalk and Barko 1985) and is refractory to decomposition. Consequences of this aging process can result in dramatic changes in the species composition of littoral macrophyte communities (Godshalk and Barko 1985).

In addition to the endogenous properties of sediment that influence fertility, sediment nutrient availability is also affected by nutrient uptake during macrophyte growth. Evidence from field studies is accumulating to suggest that rooted submersed macrophytes, even with relatively diminutive root systems, are capable of markedly depleting pools of N and P in sediments (Prentki 1979, Short 1983, Trisal and Kaul 1983, Carignan 1985). By way of confirmation, recent laboratory studies have demonstrated greater than 90- and 30-percent reductions in concentrations of exchangeable N and extractable P, respectively, from sediment over two 6-week periods of growth of *Hydrilla verticillata* (Barko et al. 1988).

As emphasized above, under most circumstances, rooted submersed macrophytes rely on sediment for a major portion of their N and P economy. Thus, even in fertile systems, depletion of sediment nutrient pools resulting from aquatic macrophyte uptake may significantly reduce sediment nutrient availability. High productivity and biomass turnover of

aquatic macrophytes in fertile systems contribute to high rates of nutrient loss from sediments (Smith and Adams 1986). Given the potential significance of sediment nutrient depletion in aquatic plant management (see below), ecological processes balancing these deficits need to be examined in detail.

Management Implications and Associated Recommendations

The sustained vigor of submersed macrophyte communities requires a balance between nutrient uptake and regeneration. Processes affecting this balance need to be considered explicitly within the context of macrophyte management. Inorganic sedimentation is frequently accelerated by human activities in the watershed, and for unknown reasons aquatic systems affected by these disturbances are often most susceptible to the invasion and subsequent explosive growth of introduced macrophyte species.

Nitrogen is a key element for the growth of rooted macrophytes. Thus, advances in our understanding of factors regulating sediment N availability are a prerequisite to the development of management approaches based on reductions in sediment nutrient availability. Toward this end, the role of submersed macrophytes in the N economy of aquatic systems needs to be investigated. A variety of physical, chemical, and biological processes (e.g., sedimentation, mineralization, and particulate movement by benthic invertebrates) that potentially contribute to sediment N availability need to be evaluated within the context of macrophyte nutrition.

Laboratory studies need to be conducted to improve our understanding of spatial aspects of sediment nutrient availability. Initially, these studies should address potential differences in the rooting depth of a variety of macrophyte species. This information will be useful in assessing the extent to which species with different rooting capacities may respond to sediment nutrient reductions. The feasibility of lessening nitrogen availability to macrophytes by interfering with naturally

occurring chemical and biological processes, thus retarding the growth potential of nuisance species, needs to be investigated. As an extension of this effort, the possibility of perpetuating reductions in nitrogen availability to nuisance species by interplanting preferred native macrophyte species should be examined.

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Rooting Depth of *Myriophyllum spicatum* L. in Relation to Sediment Nutrient Availability

by

D. G. McFarland¹ and J. W. Barko¹

Introduction

Over the past decade, considerable investigative attention has focused on the growth of submersed aquatic macrophytes relative to the chemical composition of natural sediments (for synthesis, see Barko, Gunnison, and Carpenter 1991). Presently, it is well known that, in addition to providing a medium for the attachment of rooted species, sediment provides a source of key nutrients affecting growth, such as nitrogen (N) and phosphorus (P). The availability of these and other essential sediment nutrients is affected largely by physical and chemical properties of sediment (Denny 1980; Barko and Smart 1981, 1986).

While a variety of limnological processes may modify the nutrient composition of sediment (e.g., sedimentation, diffusion, mineralization, bioturbation, etc.), one process of major importance involves direct nutrient uptake by macrophyte roots in the sediment profile (Chen and Barko 1988; Barko, Gunnison, and Carpenter 1991). Recent laboratory and field investigations have shown significant reductions in sediment nutrient reserves in the root zones of submersed macrophytes (e.g., Barko and Smart 1980; Barko 1982; Smith and Adams 1986; Barko et al. 1988; McCreary, McFarland, and Barko, in press). Results of these studies suggest that differences among species, both in accessing sediment nutrients and in adjusting to marked reductions in sediment nutrient pools, may influence the vegetational composition of littoral communities (Barko et al. 1988; Barko, Gunnison, and Carpenter 1991; McCreary, McFarland, and Barko, in press).

In this article, we present results of an investigation designed to examine the rooting capabilities of an adventive submersed macrophyte species, *Myriophyllum spicatum* L. Specific objectives of the study were to (a) determine the potential rooting depth of *M. spicatum* in nutrient-rich and nutrient-poor sediments, (b) assess changes in sediment nutrient pools within the root zones of this species, and (c) provide implications of these results for future research and aquatic plant management.

Methods and Materials

The study was conducted over a 6-week period during July-August in a greenhouse facility at the Waterways Experiment Station, Vicksburg, MS. Ten 1,200-L fiberglass tanks were filled 83 cm deep with a low-alkalinity culture solution described in Smart and Barko (1985). In general, this solution contained major nutrients except N and P, to minimize algal growth and allow sediment as the source of these nutrients for macrophyte uptake.

One liquid circulator per tank provided continuous water circulation and temperature control at 25 ± 1 °C. Maximum midday photosynthetically active radiation levels inside the tanks averaged $500 \mu\text{E}/\text{m}^2/\text{sec}$ using a neutral density shade fabric over the roof of the greenhouse that reduced natural irradiance by approximately 75 percent.

Fine-textured inorganic sediment from Brown's Lake, WES, was used to obtain two fertility levels—one provided as "used"

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sediment rendered infertile because of previous support of submersed macrophyte growth, and the other as "fresh" N-amended sediment, prepared by fertilizing with 0.8 g NH₄Cl/L wet sediment. Initial concentrations of N and other important macronutrients in the extractable and interstitial water pools are presented in Table 1. Based on these data and results of previous studies conducted in this laboratory, the N level in the unfertilized (used) sediment was acutely growth-limiting.

Table 1
Initial Sediment Nutrient Concentrations¹

Nutrient Pool	Sediment	
	Fertilized	Unfertilized
Extractable, mg/g dry sediment		
Nitrogen (NH ₄ -N)	0.32 ± 0.00	0.02 ± 0.00
Phosphorus (PO ₄ -P)	0.11 ± 0.00	0.10 ± 0.00
Potassium (K)	0.11 ± 0.00	0.10 ± 0.00
Dissolved, mg/L interstitial water		
Nitrogen (NH ₄ -N)	61.20 ± 0.61	0.92 ± 0.03
Phosphorus (PO ₄ -P)	0.80 ± 0.20	0.76 ± 0.12
Potassium (K)	11.93 ± 0.33	3.63 ± 0.03

¹ Values are means ± standard errors based on analyses of six replicate sediment samples.

Five tanks were allotted for each of the two fertility levels. Within each level, four tanks were planted with *M. spicatum*, and the remaining tank was left as a nonplanted control. Each group of planted tanks was assigned a range of sediment depths in increments of 10 cm, from 10 to 40 cm; the maximum (40-cm) sediment depth was assigned to the controls. Experimental depths were achieved by using 10-cm-diam polyvinyl chloride tube cut and capped at one end to hold the desired sediment depth. The sediment tubes were inserted individually inside supporting cylinders (15-cm-diam) to hold sediment surfaces at the same level in all tanks.

Fertility/depth treatment combinations were established separately by tank, and replicated six times in each. Separate control tanks at each fertility level were assigned three replicates apiece.

Myriophyllum spicatum used in the study was obtained from a 5-week-old greenhouse stock. Stem apices were clipped 15 cm in length and were planted six per container, with basal ends approximately 3 cm deep in the sediment.

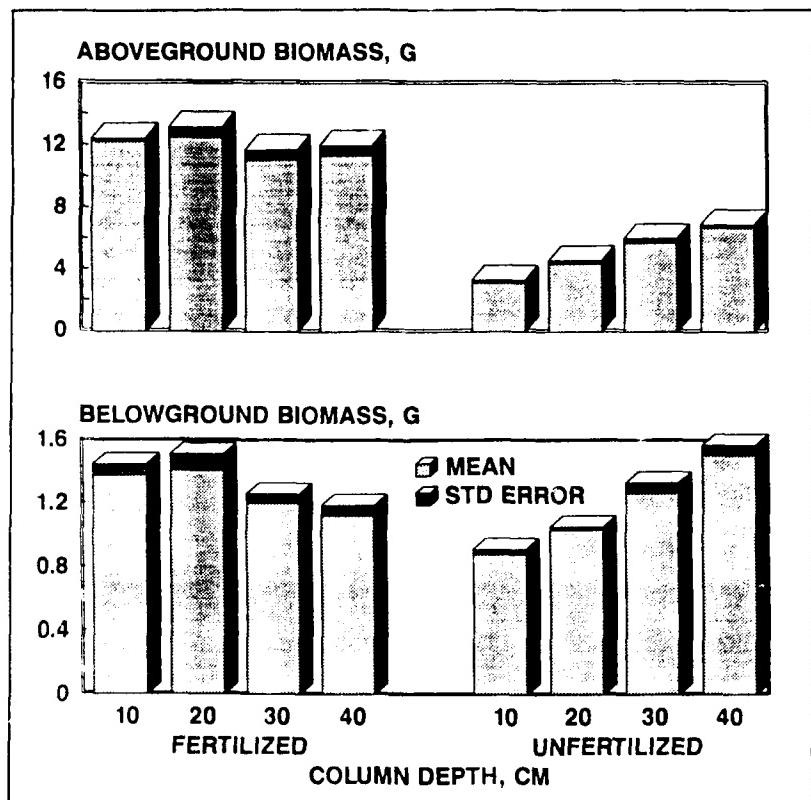
At the end of the 6-week growth period, aboveground plant material was clipped at the sediment surface and processed for dry mass and tissue nutrient determinations (procedures described in Barko et al. 1988).

Sediment, free of aboveground plant mass, was then sectioned horizontally every 5 cm from top to bottom of the sediment tube. These sections were subsequently laid flat and cross-sectioned vertically into two 202-ml halves. One half was used for sediment nutrient determinations, and the other, for belowground biomass analysis (as presented in Barko et al. 1988).

Results

Aboveground biomass production in *Myriophyllum* was strongly inhibited overall by low N availability in the unfertilized sediment (Figure 1). On fertilized sediment, aboveground biomass was about the same regardless of sediment depth. However, consecutive increases in depth of the unfertilized sediment promoted an approximately linear increase in aboveground biomass.

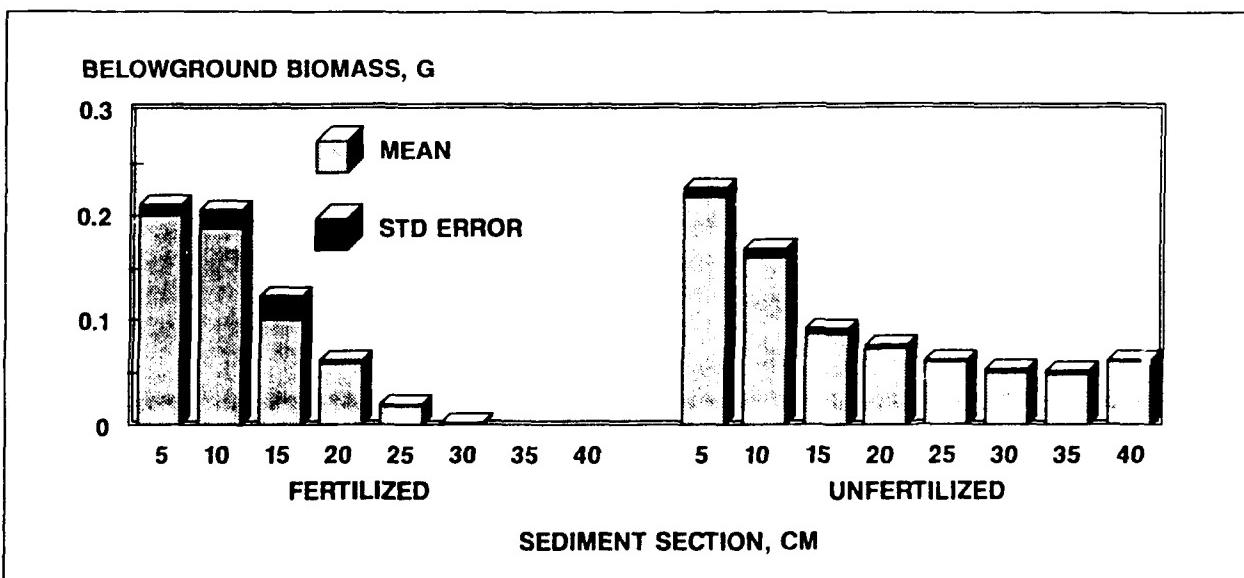
Belowground production on the fertilized sediment varied to a minor extent among sediment depth treatments (Figure 1). However, increased depth (from 10 to 40 cm) of the unfertilized sediment effected an approximate twofold increase in belowground biomass. Notably, the allocation of biomass to belowground structures was proportionately greater on the unfertilized sediment where, in corresponding depth treatments, ratios of aboveground to belowground biomass were about 2 times greater than on fertilized sediment.



*Figure 1. Aboveground and belowground biomass production in *Myriophyllum spicatum L.* planted in fertilized and unfertilized sediments of different depths. Values are means ($n = 6$) with associated standard error bars*

Sediment depth profiles down to 40 cm showed that belowground biomass in *Myriophyllum* was generally greatest within the top 10 cm (Figure 2). In the fertilized sediment, belowground biomass decreased beyond 10 cm to a minimum at 30 cm. In the unfertilized sediment, belowground biomass decreased to about 35 cm. However, because of the slight increase in biomass at 40 cm, it appears that the roots were restricted, and may have achieved greater depths than the maximum 40 cm provided in this study.

For each of the control (i.e., nonplanted fertilized and unfertilized) sediments, exchangeable N concentrations showed minimal variation along depth gradients below 5 cm (Figures 3 and 4). In most cases, however, N concentrations within the top 5 cm of sediment were



*Figure 2. Belowground biomass of *Myriophyllum spicatum L.* in consecutive 5-cm (202-ml) sections of sediment (i.e., fertilized and unfertilized), from top to bottom of 40-cm-deep sediment tubes. Values are means ($n = 6$) with associated standard error bars*

Figure 3. Final concentrations of exchangeable N determined from consecutive 5-cm sections (from top to bottom) of fertilized sediment, in 40-cm-deep sediment tubes. Means, for both non-planted control sediment ($n = 3$) and planted sediment ($n = 6$), are presented in top and bottom panels, respectively, with associated standard error bars

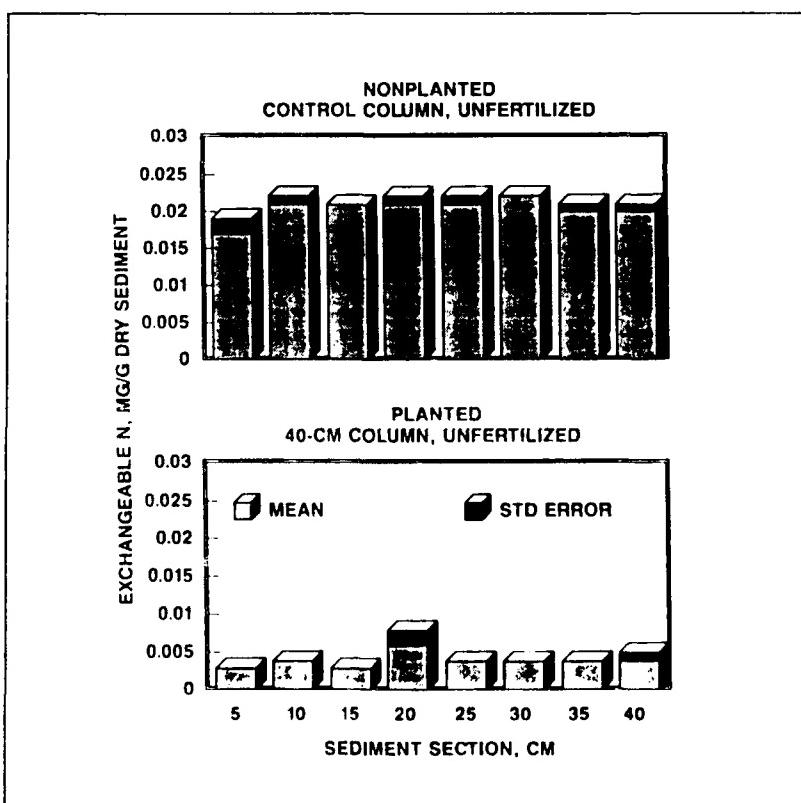
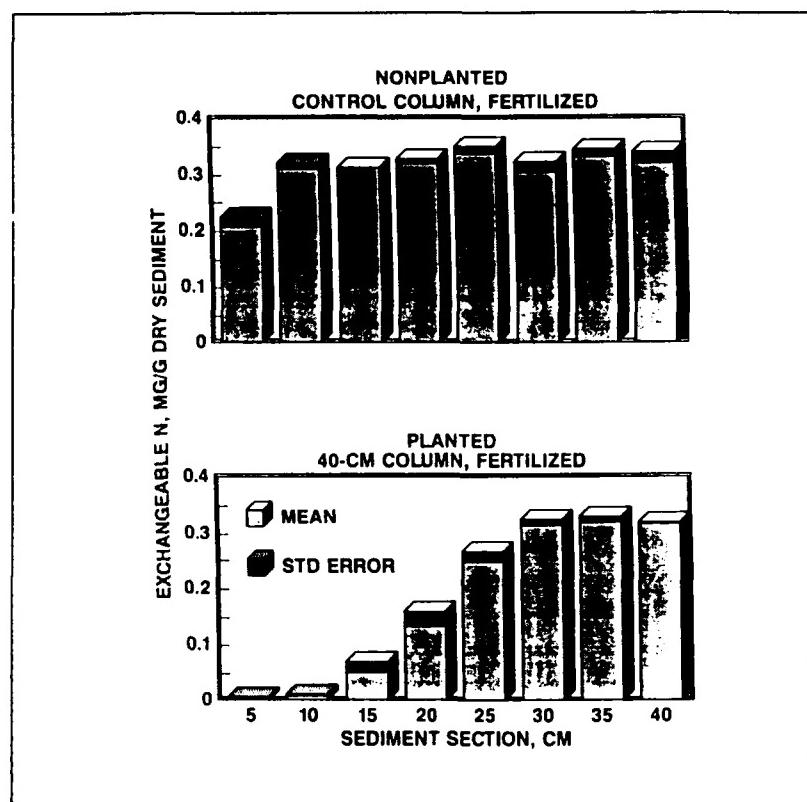


Figure 4. Final concentrations of exchangeable N determined from consecutive 5-cm sections (from top to bottom) of unfertilized sediment, in 40-cm-deep sediment tubes. Means, for both nonplanted control sediment ($n = 3$) and planted sediment ($n = 6$), are presented in top and bottom panels, respectively, with associated standard error bars

somewhat depleted, apparently due to diffusion of this nutrient into the water column.

In fertilized sediment planted with *M. spicatum*, marked reductions in exchangeable N concentrations occurred down to 25 cm, just 5 cm short of the maximum (30-cm) rooting depth achieved in this treatment (Figure 3). Here, exchangeable N reductions were greatest in the top 10 cm, and generally decreased with decreased belowground biomass (cf. Figure 2) and increased sediment depth. In direct contrast, exchangeable N in the planted, unfertilized sediment was somewhat evenly and almost completely removed along the entire 40-cm rooting depth achieved in this sediment column (Figure 4).

Discussion and Future Research

Results obtained in this investigation and elsewhere (Anderson and Kalf 1986) show that biomass production in *M. spicatum* can be strongly inhibited by low N availability in sediment. Under these conditions, *M. spicatum* allocates a substantial portion of its overall production to root formation, as evidenced by increased rooting depth (to at least 40 cm) in the present study. Increased below-ground production is characteristically noted in submersed macrophytes as the result of sediment nutrient limitation (Denny 1972, Barko and Smart 1986). This response is thought to enhance macrophyte nutrition by maximizing the volume of sediment in direct contact with root surfaces (Barko and Smart 1986).

Future research of the effects of sediment fertility, to be conducted in this laboratory, will contrast rooting depth and biomass allocation in *M. spicatum* with other important submersed macrophyte species. Such studies will increase our understanding of the relative rooting capabilities of selected species, and ultimately contribute to our ability to predict changes in submersed macrophyte community composition relative to changes in sediment nutrient availability.

As demonstrated here, growth of *M. spicatum* can result in marked reductions in sediment nutrient (i.e., exchangeable N) pools. Thus far, field investigations of N uptake by rooted submersed macrophytes have generally been quite limited (cf. Barko, Gunnison, and Carpenter 1991). However, the *in situ* studies of Prentki (1979) and Smith and Adams (1986) have shown substantial P depletion from sediments within the root zones of *M. spicatum*. A significant implication of these collective findings is that, without sufficient nutrient replenishment to sediment, growth of submersed macrophytes can be self-limiting.

Replenishment of nutrients via sedimentation is recognized as a potentially important means of counterbalancing nutrient losses in sediment due to macrophyte uptake (Barko et al. 1988). Field investigations are needed to examine, over the long term, nutrient replenishment (through sedimentation) in relation to uptake by a variety of submersed macrophyte species. This information could greatly advance our understanding of nutritional factors affecting macrophyte succession, especially periods of dominance by different macrophyte species, in littoral environments.

While results of this study show that the rooting depth of *M. spicatum* can be affected by nutrient availability in sediment, the extent to which sediment texture can influence this response is unknown. Sediment texture has been suggested to be an important factor influencing rooting depth in submersed macrophytes (Denny 1980). Differences among species in their ability to penetrate and take up nutrients in different sediment types may influence competitive relationships. Information on this aspect of submersed macrophyte ecology could be helpful in planting programs to establish native nonadventive species in areas potentially invaded by nuisance species such as *M. spicatum*.

Acknowledgments

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Aquatic Plant Competition Studies in Guntersville Reservoir

by
*R. Michael Smart*¹

Introduction

Guntersville Reservoir has been plagued with an overabundance of submersed aquatic plants for many years. *Myriophyllum spicatum* (Eurasian watermilfoil) has been the dominant weedy species thus far; however, *Hydrilla verticillata* (hydrilla) populations have been spreading recently. Although seasonal growth of both of these species can be controlled by chemical treatment, regrowth and/or reinvasion makes subsequent chemical treatments necessary. Removal of existing vegetation, by opening up potential new habitat, may actually accelerate the rate of infestation by weedy species. Once open habitats are created, weedy species (which are well adapted for rapid invasion) can extend their distribution into areas once occupied by other species.

Another major aquatic plant problem in Guntersville Reservoir is the widespread occurrence of floating mats of the filamentous blue-green alga *Lynghya wollei*. These algal mats develop on the sediment surface in shallow waters and can achieve considerable mass before floating to the water surface and becoming visible. This growth strategy allows *Lynghya* populations to achieve substantial development prior to the implementation of any management or control measures. After it is detected, *Lynghya* is difficult to control because, even after chemical treatment, the dead mats can remain intact for long periods. A more effective means for controlling *Lynghya* would be to prevent the initial subsurface development.

Lynghya problems have sometimes followed eradication of rooted submersed aquatic plants such as hydrilla. If the growth of *Lynghya* in these situations was being held in check by the rooted submersed aquatic plants, it may be possible to prevent the development of *Lynghya* by following submersed plant control efforts with establishment of rooted aquatic plants in shallow areas likely to exhibit *Lynghya* problems.

An effective management strategy for Guntersville Reservoir might be to follow the chemical removal of Eurasian watermilfoil with establishment of beneficial, nonweedy, native species that would occupy these areas to prevent reinvasion by weedy species. The objective of this research is to identify methods for promoting the successful establishment of native, nonweedy species of aquatic plants in an attempt to slow the spread of weedy species.

Past Research

As part of a major aquatic plant control effort in Guntersville Reservoir conducted jointly by the US Army Corps of Engineers and the Tennessee Valley Authority (TVA), large numbers of grass carp have been introduced into the reservoir. These fish prefer native species and hydrilla over Eurasian watermilfoil. Since the large numbers of fish could quickly consume the relatively small amounts of plant material used in experimental plots, it will be necessary to exclude the

¹ US Army Engineer Waterways Experiment Station, Lewisville Aquatic Ecosystem Research Facility, Lewisville, TX.

grass carp from the study areas, at least during the first year or two of establishment.

Murphy Hill Pond study

During the first growing season we elected to conduct our study in a pond at TVA's Murphy Hill Aquatic Research Station. This pond was located immediately adjacent to Guntersville Reservoir, exhibited almost complete coverage with a near-monotypic Eurasian watermilfoil-dominated aquatic plant community, and was expected to be free of grass carp. Conducting our initial study in the pond environment provided a learning opportunity and a chance to refine methodology prior to field testing in Guntersville Reservoir. Subsequent experiments have been conducted inside exclosures (fences) both in the pond and in the reservoir itself.

The experimental objective was to determine the effects of benthic barrier and fertilizer application on the establishment and persistence of *Vallisneria americana* and *Potamogeton pectinatus* (sago pondweed) in a water body dominated by Eurasian watermilfoil. The experimental design consisted of a factorial arrangement with two sediment treatments (barrier and no barrier), two fertility levels (control and fertilized sediments), and three planting treatments (*Vallisneria*, pondweed, and unplanted controls), and included two harvest dates for evaluating results. The experiment was replicated four times for a total of 96 experimental units or subplots.

Methods. Prior to the start of the experiment, a 1-acre plot was laid out in the middle of a large expanse of monotypic Eurasian watermilfoil. This area was treated by TVA with a granular endothall formulation. The treatment produced excellent control, providing complete kill within the plot and leaving Eurasian watermilfoil intact all around the periphery of the plot.

Each of the subplots consisted of a 1- by 1-m polyvinyl chloride (PVC) pipe frame, held in place by attachment to the anchored benthic barrier or to a rope grid laid out on

the bottom of the pond. A planting frame was constructed which divided the subplots into 36 cells approximately 15 by 15 cm. We then planted 36 bundles of approximately three to five *Vallisneria* plants or 36 sets of three sago pondweed tubers using the frame as a guide.

Results. When we returned after 5 weeks to evaluate the growth of the plants, we found that none had survived. Subsequent observations and trapping indicated that the plants had been consumed by turtles (*Chrysemys* spp.). An exclosure was subsequently constructed, and turtles were removed from within prior to replanting.

Current Research

Murphy Hill pond study

Methods. Following the construction of the exclosure, the experimental design was reduced to 48 subplots (24 on each of two benthic barrier blocks). Twelve subplots on each of the blocks were planted with *Vallisneria* in September 1990. Sago pondweed tubers were unavailable at that time, and we elected to replant with *Potamogeton nodosus* (American pondweed) tubers in the other 12 subplots of each barrier block in the spring of 1991. The plots were monitored several times during 1991.

Results. The *Vallisneria* plots were evaluated in October 1990 and appeared to be viable. Eurasian watermilfoil was beginning to regrow/reinvade the plots at that time, and this continued to occur throughout the spring and summer. Although American pondweed began vigorous growth in the spring, biomass of both species began to visibly decline during the summer months. By the end of the summer, very little *Vallisneria* remained, and both the *Vallisneria* and pondweed plots were being taken over by Eurasian watermilfoil.

The less than satisfactory results obtained in the Murphy Hill pond study can be attributed to several factors. The *Vallisneria* may have been planted too late in the season for it to become well established. Pondweed,

which was planted the following spring, was more successful in becoming established, but by this time the endemic Eurasian watermilfoil was recovering from the herbicide treatment of a year earlier. We continued to experience problems with herbivory in spite of the exclosure. Again, the herbivory was attributed to turtles, which may have entered the enclosure through holes in the deteriorating wire fencing.

The benthic barriers that surrounded the plants tended to billow out because of the buoyancy of trapped gas bubbles. Movement of the barrier, as the result of water currents or escaping gas bubbles, caused increased levels of turbidity around the plants by resuspending sediments. These suspended sediments tended to deposit on the plant leaves and may have reduced growth by limiting the amount of light available for photosynthesis. Finally, the 1- by 1-m plots may not have been large enough for optimal establishment. Larger plots would be less susceptible to herbivory and might also help the plants survive higher levels of suspended sediments by promoting deposition, thereby increasing available light levels.

Chisenhall embayment study

The objective of this study was to evaluate the abilities of *Vallisneria* and pondweed to resist invasion by Eurasian watermilfoil in Guntersville Reservoir. We selected a small cove containing a continuous bed of near-monospecific Eurasian watermilfoil located near the middle of Guntersville Reservoir.

Methods. An exclosure measuring 70 by 100 ft was constructed to exclude grass carp and other herbivores, and a herbicide treatment was applied to eliminate the watermilfoil population. We established twenty-four 5- by 5-ft experimental plots constructed of PVC pipe. Each plot was divided into one hundred 15- by 15-cm planting cells by stringing nylon cord across the plot frames. The frames were then anchored to the sediment. Experimental treatments included plantings of *Vallisneria* or pondweed (eight replicates of each species), a mixed planting of both species (four replicates), or unplanted controls (four replicates).

Treatments were randomly allocated to the plots. The plots were planted on June 3, 1991, with *Vallisneria* plants that had been obtained a few days earlier from the Holston River in Tennessee, or with tubers (winter-buds) of American pondweed collected earlier in the spring from Cedar Creek Reservoir in Alabama. Tubers had been stored moist in a refrigerator at 7 °C prior to planting. Survival of the *Vallisneria* plants was very low, and we elected to replant. A population of *Vallisneria* was located in Guntersville Reservoir, and plants were collected during the afternoon of June 27, 1991, and planted the following morning.

Results. Within 4 weeks of planting, pondweed had reached the surface of the water and was beginning to send stolons and new shoots out of the original plots. The *Vallisneria* seemed to be surviving 1 week after replanting. We began sampling biomass in late July, and continued monthly through October. The biomass of pondweed was initially high, but declined throughout the summer as it did in the Murphy Hill pond. Survival of *Vallisneria* was very low, and biomass declined from initially low levels to only a token amount. During several of the sampling trips we noticed large rafts of uprooted or torn stems of pondweed floating within the exclosure. We attributed this damage to muskrats.

***Lyngbya* study**

The objective of this study was to evaluate the establishment and growth of desirable emergent and floating-leaved species of aquatic plants in shallow-water areas that are often dominated by the mat-forming, filamentous, blue-green alga *Lyngbya wollei*. Our long-term goal is to establish desirable species of rooted aquatic plants in these areas to compete for sunlight and sediment nutrients. By reducing levels of both of these resources, we hope to reduce the magnitude of *Lyngbya* and other aquatic plant problems in shallow water.

Methods. We selected four emergent and two floating-leaved species, based on their

desirability for creating habitat, ease of control or lack of problem-causing potential, expected ability to grow in *Lyngbya*-dominated areas, and occurrence in Guntersville Reservoir. The species selected (all native to the United States) included *Eleocharis quadrangulata* (spikerush), *Justicia americana* (American waterwillow), *Nelumbo lutea* (American lotus), *Potamogeton nodosus* (American pondweed), *Saururus cernuus* (lizard's tail), and *Scirpus validus* (softstem bulrush).

We selected two *Lyngbya*-dominated sites (Ossa-Win-Tha and Waterfront) and a control site (Boshart Creek). Water depths at each site were similar, ranging from 1.7 to 2.5 ft at Ossa-Win-Tha, 1.7 to 2.7 at Waterfront, and 1.8 to 3.0 at Boshart Creek. At each site we constructed a 32- by 64-ft exclosure to exclude grass carp and other herbivores. We established fourteen 4- by 8-ft experimental plots constructed of PVC pipe. Each plot was divided into thirty-two 1- by 1-ft planting cells or eight 2- by 2-ft planting cells (American lotus) by stringing nylon cord across the plot frames. The frames were then anchored to the sediment.

Experimental treatments included two replicate plots of each species as well as two replicate control (unplanted) plots. Plant species were assigned to blocks along the depth gradient. Spikerush and lizard's tail were assigned to the shallowest depth block; bulrush and waterwillow were assigned to intermediate depths; and lotus, pondweed, and the control plots were assigned to the deepest block. Plots were planted on June 26 and 27, 1991, with transplants of each of the species except pondweed. Transplant stock had been obtained a few days earlier from locations within Guntersville Reservoir. Pondweed was planted from tubers (winterbuds) that had been collected in March from Cedar Creek Reservoir in Alabama. Tubers had been kept moist in a refrigerator at 7 °C prior to planting.

The performance of the plants was evaluated every 2 weeks during the growing season. Parameters considered included percent survival and percent cover of the planted species, and percent cover of undesirable species (*Lyngbya* or Eurasian watermilfoil). We also collected water samples every 2 weeks for analysis of nutrients.

Results. All of the American lotus transplants died very soon after planting, and we elected to replant this species with seed. All of the other species exhibited at least some survival, at least at the control site. By the end of September 1991, the species had grown to near their maximum for the season. All of the transplanted species exhibited nearly 50 percent or greater cover at the control site (Figure 1), and American lotus grown from seed had achieved over 25 percent cover at this site. Spikerush and pondweed exhibited the best performance, achieving 90 percent cover at the control site by the end of September.

Results differed at the *Lyngbya*-dominated sites. Spikerush and bulrush failed to establish at either Ossa-Win-Tha or Waterfront (Figure 1). American waterwillow and American lotus survived at Ossa-Win-Tha but not at Waterfront. Lizard's tail and American pondweed survived at both sites. Of all the species evaluated, American pondweed exhibited the greatest potential for competing with established *Lyngbya*. This species achieved 90 percent cover at both *Lyngbya*-dominated sites and at the control site.

Conclusions

Vallisneria has been difficult to establish in both the Murphy Hill pond and in Chisenhall embayment in Guntersville Reservoir. This species is difficult to transplant, particularly in soft, low-density sediments such as are found in both of these sites. Better success has been obtained using dormant propagules such as winterbuds.¹ Future work on this

¹ Carl E. Korschgen and W. L. Green. 1988. American wildcelery (*Vallisneria americana*): Ecological considerations for restoration. Technical Report 19. Washington, DC: US Department of the Interior, Fish and Wildlife Service.

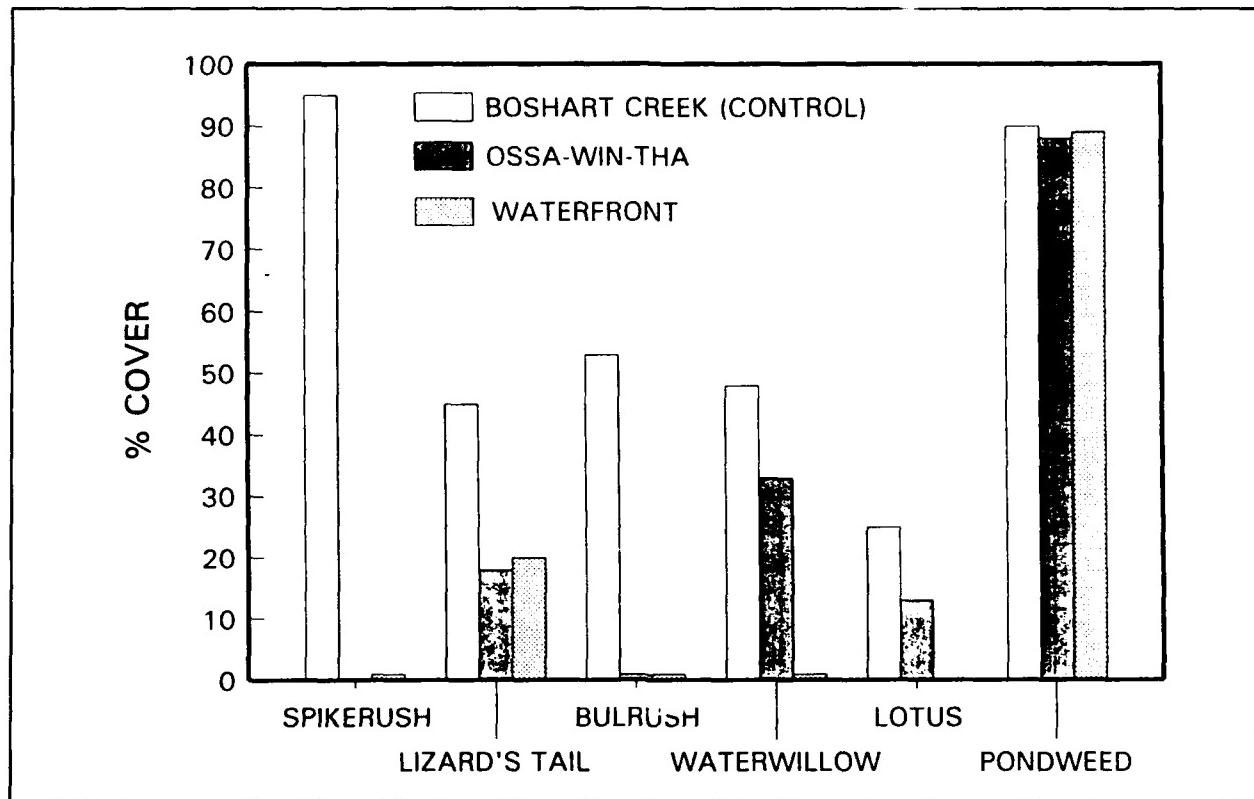


Figure 1. Percent cover of selected species in *Lyngbya*-dominated (Ossa-Win-Tha and Waterfront) and control (Boshart Creek) sites, September 1991

species will employ winterbuds rather than transplants wherever possible.

American pondweed is easily established using winterbuds, and this species grows rapidly once established. However, both pondweed and *Vallisneria* are susceptible to herbivory, and this remains a severe impediment to the successful establishment of these desirable native species in Guntersville Reservoir. Overgrazing is most damaging during the early establishment phase. At this time there is very little vegetation present, and its ability to recover from grazing is minimal. Herbivore exclusion will be required to establish desirable submersed aquatic plants in most systems.

In addition to the direct effects exerted by the grass carp through overgrazing preferred plant species, it is likely that grass carp also exert significant indirect effects. By substantially reducing the overall level of submersed

aquatic vegetation in the lake as well as by changing the species composition of the vegetation, grass carp may affect the foraging activities of other herbivores in the system. Although we are able to exclude grass carp from our exclosures, grazing pressure from other herbivores may actually increase as food becomes less available outside the exclosures. This indirect effect of grass carp deserves further study.

Several native plant species exhibited some potential for competing with *Lyngbya* and other problem aquatic plants in shallow water. The presence of desirable emergent or floating-leaved vegetation in these shallow waters should reduce populations of *Lyngbya* and other weedy aquatic plant species in these areas, thus reducing their potential to spread or reinfest other sites by fragmentation. By promoting the establishment of desirable rooted vegetation in shallow water we may also be able to reduce nutrient export from

these areas into deeper water, thereby lessening aquatic weed problems elsewhere. These aspects of this work should be considered in further studies.

Future Research

Murphy Hill pond study

In fiscal year 1992 we will repair the enclosure, remove the benthic barrier, repeat the herbicide treatment to eliminate existing Eurasian watermilfoil, and replant *Vallisneria* and pondweed. Plot sizes will be increased from 1 by 1 m to 5 by 5 m, and we will add another species, American lotus. Planting will be accomplished in the spring of 1992.

Chisenhall Embayment study

We will replant the harvested pondweed plots and all *Vallisneria* plots and will continue sampling established plots and control plots. *Vallisneria* plots will be planted with winterbuds rather than transplants. We will be more vigilant at trapping muskrats in order to minimize herbivory.

***Lyngbya* study**

We will replant plots as needed to make up for the mortality that occurred last season. We will continue plot evaluations throughout the growing season.

Macrophyte-depth interaction study

We plan to initiate a study to look at optimum depths for establishing the six species used in the *Lyngbya* study. We also plan to include *Pontederia cordata* (pickerel weed) in this study. The study will be conducted at several sites in Guntersville Reservoir.

Lotus-watermilfoil study

We will initiate studies to examine competitive interactions between American lotus and Eurasian watermilfoil. In this study we will concentrate on competition between these two species for light and sediment nutrients. Preliminary studies will be conducted in experimental ponds at the Lewisville Aquatic Ecosystem Research Facility in Lewisville, TX.

Acknowledgments

Many people have contributed to the development of this research project. The contributions of the following, in particular, are gratefully acknowledged: Leon Bates, David Brewster, Earl Burns, Mark Dowdy, Robert Doyle, Susan Dutson, Larry Dyck, Aleida Eu-banks, Terry Freeman, Tammy Hancock, David Honnell, Jim Luken, Pete Mangum, John Madsen, Doug Murphy, Wayne Poppe, Joe Snow, and David Webb.

Competitive Interactions Among Introduced and Native Species

by
R. Michael Smart¹

Introduction

Excessive growths of submersed aquatic plants are largely constrained to environments featuring both a high growth potential and the presence of an exotic, weedy species. Under unfavorable growth conditions, populations of weedy species do not develop sufficiently to cause problems, and even under favorable conditions, the growth of native species does not usually reach problem proportions.

Two of the most problematic of the introduced, weedy species are *Hydrilla verticillata* (hydrilla) and *Myriophyllum spicatum* (Eurasian watermilfoil). These species are characteristic of weedy species in general. Many of the problems associated with the growth of these weedy species are associated with particular biological characteristics such as growth form. Weedy species generally develop a dense mat, or canopy, at the water surface. This surface mat causes a variety of problems, including diminished exchange of gases, water, dissolved substances, and organisms, interference with navigation and other water use, degraded water quality, and reduced habitat diversity.

Many native, nonweedy species, however, exhibit a different growth form. In these species, typified by *Vallisneria americana*, biomass is more uniformly distributed throughout the water column. Since these species do not develop an extensive surface mat, they generally do not cause significant aquatic plant problems.

The objective of this work unit is to evaluate methods for promoting the establishment and persistence of native, nonweedy species in an attempt to slow the spread of exotic, weedy species.

Approach

Small-scale, short-term studies are useful for screening possible competitive species, for identifying important mechanisms involved in competitive interactions, and for studying particular ecophysiological processes. These types of studies produce rapid results, are relatively inexpensive, and serve to focus longer term research projects. Unfortunately short-term studies provide only a brief glimpse at competitive interactions occurring during particular time periods, usually during the early stages of colonization and establishment. Information obtained in these types of studies is not easily extrapolated to larger spatial and longer time scales. Long-term, large-scale studies, on the other hand, are difficult to conduct and to control, are quite expensive, and require a long period of time to produce results. However, these types of studies provide a truer picture of the outcome of competition over meaningful spatial and temporal scales.

In consideration of the above, we have chosen a dual research approach involving study at two spatial and temporal scales:

- Short-term, small-scale studies of competitive interactions under closely controlled conditions in greenhouse tanks.

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- Longer-term observation of competition among populations growing in experimental ponds.

This dual approach will allow us to evaluate competitive species, identify mechanisms of competition, and develop hypotheses in short-term greenhouse testing and to evaluate our findings in pond trials. We consider this type of approach to be the most efficient use of temporal, physical, and financial resources.

Past Research

Small-scale studies

In a greenhouse tank experiment, we examined competition between the native species, *Vallisneria*, and the exotic species, hydrilla. The objective of the research was to identify potentially important factors and mechanisms involved in short-term competitive interactions between these species.

Methods. To elucidate factors involved in competitive interactions between these species, we conducted the experiment in a factorial arrangement with two levels of inorganic carbon availability and two levels of sediment fertility. The experiment was conducted in 1,200-L fiberglass tanks in a greenhouse facility at the Waterways Experiment Station. The two levels of carbon supply were achieved by varying the concentration of CO₂ (ambient and 10x) in the aerating gas supplied to twin airlifts in each tank. The two fertility levels were achieved by using fresh Brown's Lake sediment and the same sediment after a prior period of submerged plant growth. Each tank contained forty 1-L containers planted with four hydrilla plants, four *Vallisneria* plants, or two plants of each species. After 8 weeks, we harvested 10 replicate containers for determination of shoot and root biomass.

Results. In the absence of competition, hydrilla was more responsive to an increase in carbon supply than to an increase in nutrient availability (Figure 1). In contrast, *Vallisneria* responded to an increase in sediment fertility

but not at all to increased carbon supply. Maximal growth of both species was obtained at the high levels of both carbon supply and sediment fertility (Figure 1).

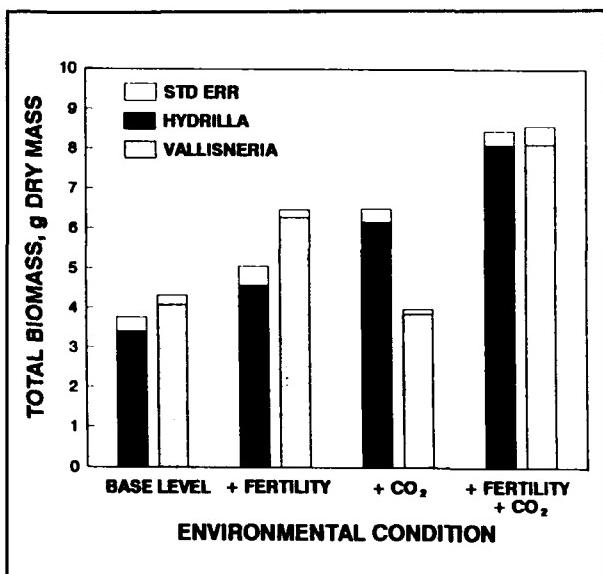


Figure 1. Total biomass responses of *Hydrilla verticillata* and *Vallisneria americana* growing monospecifically (without competition) in relation to sediment fertility and CO₂ supply. Bars represent means plus 1 standard error of the mean based on 10 replications

Since the individual species responded differently to these two environmental factors, it is not surprising that the outcome of competition between them depended on environmental conditions. Under low levels of inorganic carbon supply and sediment fertility, the two species were fairly evenly matched, each interfering with the other approximately equally (Figure 2). However, under elevated CO₂ supply at low fertility, the growth of hydrilla was more rapid than that of *Vallisneria*, and hydrilla was the superior competitor. Under the low CO₂ supply at the high-fertility condition, *Vallisneria* was more competitive. Under high levels of both factors, the two species were again evenly matched.

These results indicate that under certain conditions, *Vallisneria* can be an effective competitor with hydrilla. Increased sediment fertility in this experiment allowed the slower

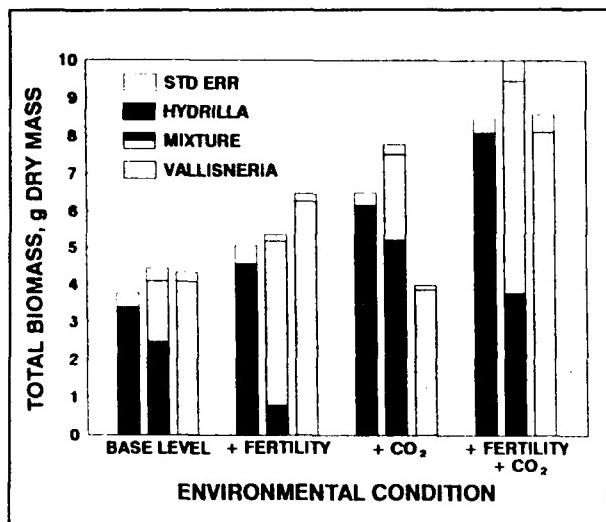


Figure 2. Total biomass responses of *Hydrilla verticillata* and *Vallisneria americana* growing together in mixture relative to biomass of the species when grown separately (without competition) in relation to sediment fertility and CO₂ supply. Bars represent means plus 1 standard error of the mean based on 10 replications

growing *Vallisneria* to become established, even in the presence of competing hydrilla. Once *Vallisneria* is successfully established, it is able to persist and even suppress the growth of hydrilla, at least over short time scales.

Large-scale studies

The experimental objective of the large-scale study was to determine the abilities of populations of the native species *Vallisneria* and *Potamogeton pectinatus* (sago pondweed) to resist invasion by hydrilla. The experiment required observation of plant responses within permanent plots over an extended period. A 1-acre pond at the Lewisville Aquatic Ecosystem Research Facility (LAERF) was selected for the study.

Methods. The plots were laid out as a series of 96 hexagonal cells in a 1-acre pond. Species were assigned to cells in a regular pattern so that each species was surrounded by three cells of each of the other two species. The sediment in every other cell of each

species was fertilized with nitrogen (ammonium sulfate), which has been shown to potentially limit growth in these ponds. The entire pond bottom was rototilled to incorporate the nitrogen fertilizer and to facilitate planting. Plants of *Vallisneria* and dormant winterbuds of sago pondweed were planted in August on 30-cm centers as the pond was being filled.

Results. Endemic submersed macrophyte species, *Najas guadalupensis* and *Chara vulgaris*, grew rapidly from a seed/spore bank in the pond sediment, overgrowing the planted native species. The growth of these two species interfered with the establishment of *Vallisneria* and sago pondweed. By October, when the pond was drained for observation and sampling, hydrilla had grown very well, attaining full coverage in the cells where it had been planted. However, hydrilla did not appreciably invade adjacent cells vegetated primarily with *Najas* and *Chara*, indicating that preemptive establishment of native vegetation might be effective at slowing the spread of hydrilla.

Current Research

Given adequate sediment fertility, *Vallisneria* can effectively outcompete hydrilla. In order to increase *Vallisneria*'s competitive advantage, we are interested in determining the possible benefits to be derived from an initial period of preemption of sediment nutrients by *Vallisneria*. Research to address this question was again initiated at two scales.

Small-scale studies

The experimental design includes two sets of treatments—one in which both species were planted at the same time (without pre-emption) and one in which *Vallisneria* is allowed a 4-week period of preemption. Each set includes the two species growing alone and two types of mixtures—a treatment with each of the species in separate containers and a treatment with both species growing together, rooted in the same sediment (Figure 3). This allows us to separately examine the importance of competition for sediment nutrients

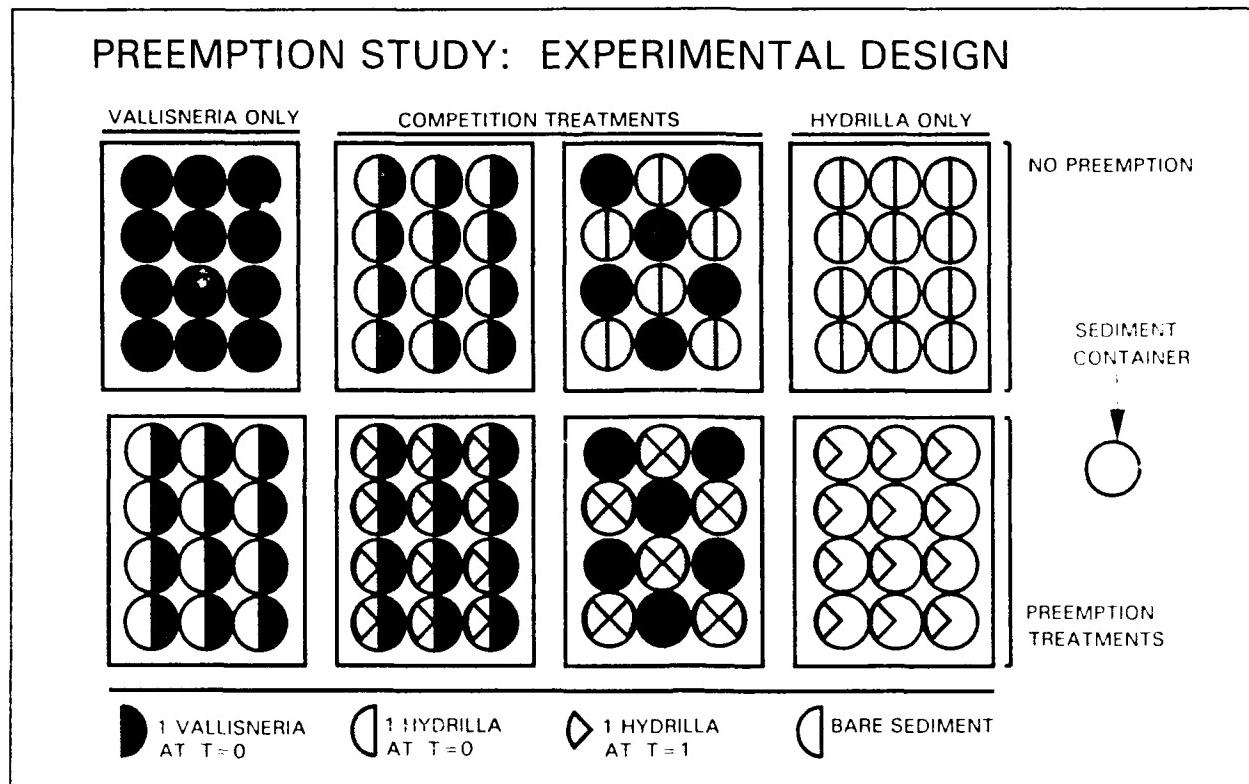


Figure 3. Diagrammatic representation of experimental design employed for evaluating preemption as a factor in competition between *Hydrilla verticillata* and *Vallisneria americana*. T_0 represents the beginning of the experiment, and T_1 represents the midpoint (T_0 plus 4 weeks). All *Vallisneria* was planted at T_0 . *Hydrilla* was planted at T_0 (direct competition) or at T_1 , after a 4-week period of preemption by *Vallisneria*. All treatments were evaluated by harvesting 12 replicate containers

in relation to overall competition. In the pre-emption portion of the experiment, we examine the growth of *Vallisneria* alone and as affected by hydrilla invasion. We also examine the growth of invading hydrilla as affected by the presence or absence of established *Vallisneria*.

Methods. The experiment was conducted in 1,200-L fiberglass tanks in a greenhouse facility at the LAERF. The experiment was conducted at high levels of both inorganic carbon supply (10 \times ambient CO₂) and high sediment fertility (fresh Lewisville pond sediment). Based on earlier studies, these conditions were not expected to selectively favor either of the two species. Each tank contained fifty-four 1-L containers planted with hydrilla and/or *Vallisneria* plants, as shown in Figure 3. After 10 weeks, we harvested 12 replicate

containers for determination of shoot and root biomass.

Results. Data analysis is not yet complete. Preliminary observations suggest that, while preemption can convey competitive benefits to *Vallisneria*, a 4-week period of preemption may be insufficient to provide a lasting advantage.

Large-scale studies

The experimental objective of the second pond study was similar to the first—to determine the abilities of populations of native species to resist invasion by hydrilla. In this experimental design, however, we included an initial period of preemption. *Vallisneria* and *Potamogeton nodosus* (pondweed) were the native species selected for this study.

The experiment again required observation of plant responses within permanent plots over an extended period.

Methods. The plots were laid out in a manner identical to that described above. Species assignments and fertilizer applications were likewise identical to the first experiment. Prior to planting the experiment, the entire pond bottom was rototilled and treated with metam-sodium to reduce numbers of spores and seeds of endemic populations of *Chara* and *Najas*, respectively. The metam-sodium treatment was also expected to eliminate tubers of hydrilla remaining from the previous experiment. Plants of *Vallisneria* and dormant winter-buds of pondweed were planted in June 1991 on 30-cm centers as the pond was being filled. Cells to be planted with hydrilla were covered with a geotextile landscaping fabric to prevent the growth of vegetation in these cells. Hydrilla will be planted after a 1-year period of preemption.

Results. Although we have observed growth of all three unwanted species (*Najas*, *Chara*, and hydrilla), the pretreatment with metam-sodium was effective at delaying the problematic growth of the endemic species. Growth of the unwanted species is being controlled by hand removal. Both *Vallisneria* and pondweed have become well established, and hydrilla will be introduced during the summer of 1992.

Development of propagation methods

Pond-scale studies require thousands of propagules of native plant species. Unfortunately many of these species are not readily available outside of the state of Florida. Since aquatic plants, like their terrestrial counterparts, may be genetically adapted to their particular geographic region, it is desirable in revegetation efforts to use planting stock that originated from within the local area. For

these reasons we have begun to study methods of aquatic plant propagation. Economical techniques of plant production will be required if competitive replacement is to be practiced on a large scale. Progress in this area is reviewed in another paper.¹

Future Research

Small-scale studies

In fiscal year 1992 we will complete analysis of the greenhouse preemption study and initiate several additional small-scale, short-term studies. These types of studies provide much valuable scientific information and also help to direct our longer term efforts. We will also continue to develop efficient methods for production of propagules of native aquatic plant species.

Intermediate-scale studies

We will initiate an intermediate-scale, seasonal study of competition using containers of plants placed in a pond environment. This type of study will allow an examination of the importance of seasonal events and phenological stage of development in affecting the outcome of competition. Specifically, we are interested in the effects of different physiological mechanisms of perennation (over-wintering), such as the onset and duration of the dormant period, the ability to sequester reserves in dormant tissues, etc.

Large-scale studies

We will continue the long-term pond study with the introduction of hydrilla to the now bare cells. We will observe the spread of hydrilla throughout the bare cells and will measure its ability to invade adjacent vegetated cells. These observations will allow determination of the abilities of *Vallisneria* and American pondweed populations to resist invasion by hydrilla. Close observation of

¹ S. E. Monteleone and R. M. Smart. 1992. See pp 195-199 in this proceedings.

these experimental populations may reveal important competitive attributes or mechanisms of invasion. These attributes or mechanisms can then be carefully studied under more controlled conditions.

Reservoir-scale studies

We have located adjacent monospecific beds of *Vallisneria* and hydrilla in a reservoir in east Texas. We will establish some permanent plots within these naturally occurring populations in order to monitor any changes in their boundaries or in species composition of the community. We also hope to initiate a field test employing preemptive establishment of beneficial, native species in a reservoir

that is not currently experiencing aquatic plant problems. This approach may be very successful in delaying the occurrence of major aquatic plant problems in new Corps of Engineers' construction projects.

Acknowledgments

Many people have contributed to the development of this research project. The contributions of the following, in particular, are gratefully acknowledged: Michael Crouch, Gary Dick, Tammy Hancock, David Holland, David Honnell, John Madsen, Kimberly Mauermann, Susan Monteleone, and Joe Snow.

Submersed Macrophyte Invasions and Declines

by
Craig S. Smith¹

Introduction

When exotic submersed macrophytes invade new locations, they often exhibit a characteristic pattern of explosive growth to a relatively stable plateau, followed by a noticeable decline (Figure 1). For Eurasian watermilfoil (*Myriophyllum spicatum* L.), the period of maximum abundance typically lasts approximately 10 to 15 years (Carpenter 1980). The maximum abundance attained by *M. spicatum* varies among water bodies, as the result of factors that are at best superficially understood (see Smith and Barko 1990,

population control mechanisms. Once these population controls have been identified, management strategies can be designed to act in concert with them. The goal of the newly initiated research on submersed macrophyte invasions and declines is to identify those factors determining both invasion success and declines, and to elucidate associated causal mechanisms.

Approach

The investigation of invasions and declines will proceed through the following stages:

- Literature review.
- Workshop.
- Construction of an invasion and decline database.
- Evaluation of increasing, stable, and decreasing plant populations.
- Experimental manipulation of proposed population control factors.
- Development of management techniques and recommendations.

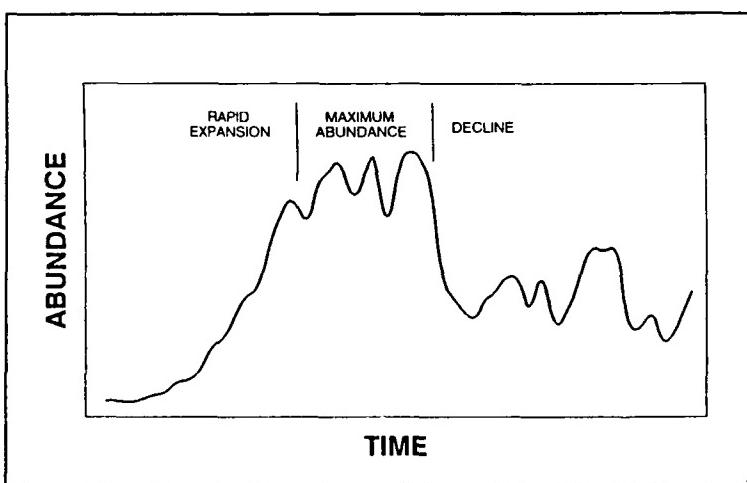


Figure 1. Typical pattern of invasion and decline when Eurasian watermilfoil invades a new location

Smith 1991). Throughout the period of maximum abundance, year-to-year increases and decreases in abundance may be substantial (Figure 1).

Fluctuations in submersed plant abundance are often evidence of the existence of natural

Initial research will focus on Eurasian watermilfoil, because its invasions and declines are the best documented to date. The mechanisms controlling Eurasian watermilfoil are probably the same as or similar to those controlling other exotic submersed macrophyte species.

¹ US Army Engineer Waterways Experiment Station, Vicksburg, MS.

Literature review

Much of the literature on factors influencing Eurasian watermilfoil invasions and declines was reviewed recently by Smith and Barko (1990). They identified several general factors influencing invasion success (Table 1) and listed proposed causes for declines (Table 2). Additional literature review will focus on cataloging information on invasions and declines of other submersed aquatic macrophyte species.

Table 1
Factors Influencing the Success of Eurasian Watermilfoil Invasions

Factor	Influence on Eurasian Watermilfoil Success
Fertility	Nuisance growths of the plant are primarily restricted to fertile lakes, or fertile locations in less fertile lakes.
Disturbance	Invasion is facilitated when disturbances open up habitat.
Human activities	Human activities spread the species between lakes.

Table 2
Proposed Causes of Eurasian Watermilfoil Declines¹

Nutrient depletion
Toxin accumulation
Shading by phytoplankton or attached algae
Parasite(s) or pathogen(s)
Harvesting/herbicides
Climatic fluctuations
Competition from other macrophytes
Insect herbivory (Painter and McCabe 1988)

¹ After Carpenter (1980) except as noted.

Workshop

A workshop will be held to discuss factors influencing invasions and declines and to collect information about submersed macrophyte population dynamics. The workshop will be held in conjunction with the 32nd Annual Meeting of the Aquatic Plant Management Society and International Symposium on the Biology and Management of Aquatic Plants, to be held in Daytona Beach, Florida, in July 1992. Experts on submersed macro-

phyte ecology from throughout North America will be invited to attend.

Invasion and decline database

Reports from aquatic plant managers around the country and from the published literature will be collected into a database of information on invasions and declines. The database will include information on when and where submersed macrophytes are invading and declining, and ancillary information concerning particular features of these events. Information in the database will be used to characterize invasions and declines and to select field sites for the study of invasions and declines.

One reason the causes of natural declines have remained elusive is that there are probably several types of declines, produced by distinct causes. Presumably, declines produced by different causes will have differing characteristics. Characterization of declines, according to features such as those listed in Table 3, will provide clues to the identity of the causative factors. For example, climatic phenomena will likely produce roughly simultaneous declines of several species over entire regions. Effects of herbivores and diseases may vary in species-specificity and geographical scope, but will likely spread outward from an epicenter.

Table 3
Characterization of Declines

Extent	Duration	Scope
One location	Intra-annual	One species
Local	Inter-annual	Many species
Regional	Several years	Whole type
	Long term	Several types

Evaluation of plant population trends

Sites having increasing, stable, and declining populations of submersed macrophytes will be examined. At each site, environmental conditions and plant status will be evaluated. Environmental measurements will include such variables as turbidity, temperature,

sediment composition (e.g., organic content, density, and nutrient content), and the presence and status of other macrophyte species nearby. Plants will be examined for herbivorous insects, pathogens, and dense growths of epiphytes. Plant vigor will be evaluated by measuring in situ rates of physiological processes, such as photosynthesis. Sediments will be collected from selected areas and returned to the laboratory, where they will be bioassayed for their ability to support plant growth. Plants from selected locations will be returned to the greenhouse for growth rate comparisons under standard conditions.

Experimental manipulation of proposed population control factors

Once the studies described above have implicated particular factors as possibly limiting the success of invasive macrophyte species, experiments will attempt to duplicate their action. Results of experimental manipulations will separate causes of poor plant performance from factors serendipitously associated with poor performance or resulting from it. Additional experiments will evaluate ways of manipulating key factors to reduce plant performance in situ.

Benefits/Products of This Research

An understanding of natural population controls will provide a basis for the development of more efficient macrophyte management strategies. Knowing how natural controls operate will make it possible to identify conditions under which management is most necessary, when it is most likely to

succeed, and when specific management techniques are likely to be inefficient or counterproductive.

It may also be possible to develop "ecological management techniques," i.e., techniques for manipulating environmental conditions to achieve plant management objectives. Several such strategies are currently under investigation elsewhere in the Aquatic Plant Control Research Program. Identification of mechanisms of natural population control will pinpoint those environmental manipulations with the greatest likelihood of success. Development of management strategies that hasten natural declines may be possible.

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Value of Aquatic Macrophytes for Invertebrates: Studies Conducted in Lake Seminole, Florida and Georgia

by

Andrew C. Miller,¹ Richard Peets,² and David C. Beckett²

Introduction

Macrophytes affect limnological processes in both lentic and lotic habitats (Carpenter and Lodge 1986). Aquatic macrophytes were shown to affect flow, water temperature, and clarity in the Potomac River in Maryland (Carter et al. 1988). Aquatic macrophytes also increase sediment deposition and substrate stability (Carpenter and Lodge 1986, McDermid and Naiman 1983). Dissolved oxygen in benthic sediments can be increased by the root systems of macrophytes. Decayed macrophytes are an important source of detritus and nutrients to benthic invertebrates (Carpenter and Lodge 1986).

Macrophytes can also affect benthic invertebrate abundance and density. Engel (1988) showed that aquatic macrophytes increased benthic invertebrate abundances in Lake Halverson, Wisconsin; over 75 percent of benthic organisms in the lake were collected beneath macrophyte beds. Beckett, Aartila, and Miller (1992a) reported that benthic densities were seven times greater in vegetated sediments in Eau Galle Lake, Wisconsin, as compared with nonvegetated sediments.

Aquatic macrophytes provide additional colonizable substrate in the littoral zone besides that afforded by the bottom. This substrate is often more complex than the homogenous benthic sediments. Krecker (1939), Rosine (1955), Gerking (1957), Soszka (1975), and Engel (1988) reported that macrophytes supported more diverse invertebrate assemblages than did adjacent ben-

thic habitats. Krecker (1939), Rosine (1955), and Rooke (1984) reported that macrophyte species with complex morphological growth patterns can support high invertebrate densities because of increased surface area. In addition to macroinvertebrates, macrophytes are often colonized by bacteria, diatoms, and algae. Invertebrate taxa feed by scraping attached organisms, or those that are filter-feeders attach to macrophytes. Predacious groups such as the Odonata and Hemiptera cling to macrophytes while searching for prey (McDermid and Naiman 1983).

The purpose of this research was to examine density and community composition of invertebrates on three species of aquatic macrophytes (*Hydrilla verticillata*, *Nymphaea odorata*, and *Potamogeton nodosus*) in Lake Seminole, Georgia and Florida.

Study Area

Lake Seminole, located in southeastern Georgia and northwestern Florida, was created in 1954 by construction of Jim Woodruff Lock and Dam at the confluence of the Flint and Chattahoochee Rivers (US Army Engineer District, Mobile 1972) (Figure 1). At the normal pool elevation of 23.5 m, the total watershed measures 15,297.1 ha. A large percentage of the lake is 2.1 m deep or less. The watershed extends 80.5 km up the Chattahoochee River and 75.6 km up the Flint River. The climate in the study area is mild; annual temperature is 20 °C. The average growing season lasts from the middle of March to the end of October.

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² University of Southern Mississippi, Department of Biology, Hattiesburg, MS.

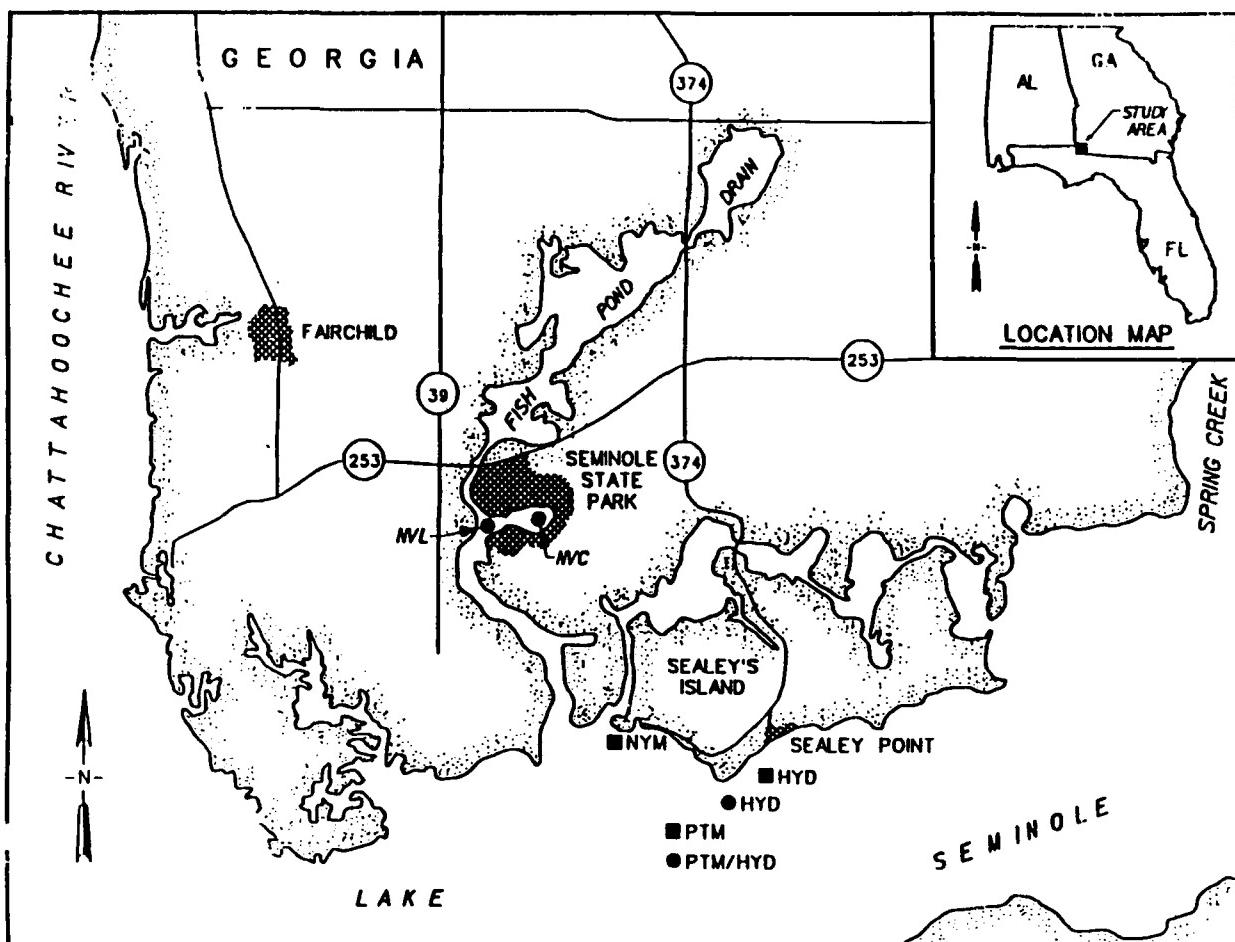


Figure 1. Map of the study area

As of 1990, Lake Seminole contained approximately 220 species of macrophytes and three species of algae. Macrophytes covered approximately 60 percent of the lake (Joseph Kight, personal communication).

Methods

Macrophytes were collected in Lake Seminole, Georgia, on July 12, 1987. Each of the three species was taken at three replicated sites. Five samples were taken in each site; a total of 15 plants were collected per species. For *N. odorata* and *P. nodosus*, a sample consisted of an entire plant (excluding any portions in the sediments). These plant species were snipped at the substrate-water interface. Since *H. verticillata* grows in tangled masses, it was not possible to determine the extent of a single plant. Therefore, for *H. verticillata*, a single section of stem was cut with scissors

and placed in a container with 5-percent formalin solution.

Macrophytes were preserved in the field with 5-percent formalin and transported to the University of Southern Mississippi, where they were stained with Rose Bengal to aid in invertebrate identification (Mason and Yevich 1967). In the laboratory, plants were rinsed through a 250- μm sieve. Invertebrates were picked from individual plants with the aid of a dissecting microscope and preserved in 75-percent alcohol. Chironomids and oligochaetes were mounted for identification following the procedure of Beckett and Lewis (1982).

Results

A total of 16 taxonomic groups were identified on three species of macrophytes in

Lake Seminole. Fourteen taxonomic groups were common to the stems and leaves of these three macrophyte species and the benthic sediments. Hydracarina and the microcrustaceans Copepoda, Ostracoda, and Cladocera were common to both habitats. Two taxa of Coleoptera were present on macrophytes compared with only one taxon from the sediments. Diptera and Oligochaeta were common in sediments and on plants. Two groups (Bivalvia and Amphipoda) were found only in sediments, and two groups (Lepidoptera and Cnidaria) were unique to the macrophytes.

Total density (mean of three sites) was greatest on *H. verticillata* (12,843 individuals/sq m), moderate on *P. nodosus* (9,689 individuals/sq m), and least on *N. odorata* (5,922 individuals/sq m) (Figure 2).

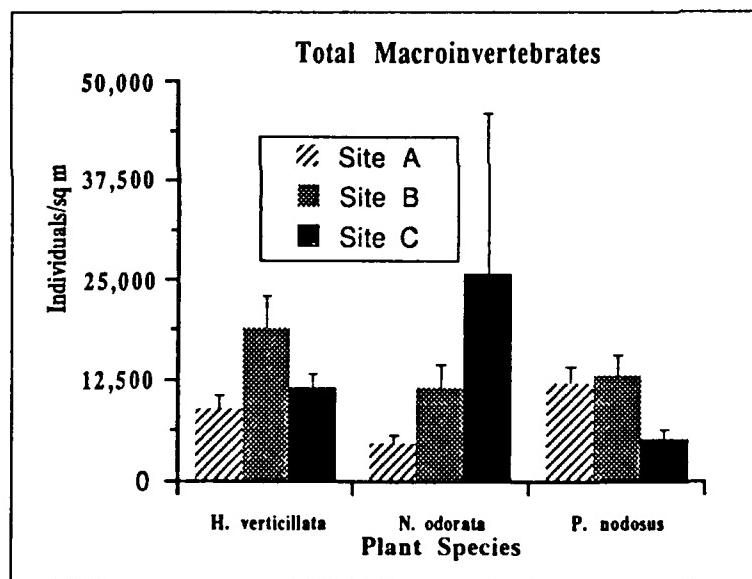


Figure 2. Mean density (and standard error bars) of total macroinvertebrates collected on three plant species ($N = 5$)

The relationship between total number of individuals collected and total stem and leaf surface area was examined for the three macrophyte species. For total macroinvertebrates, there was a weak positive relationship for *H. verticillata* and *P. nodosus*, but virtually no correlation for *N. odorata* (Figure 3). Only two taxa (Oligochaeta and Nematoda)

and total macroinvertebrates had a positive correlation between abundance and surface area for all three macrophyte species. Copepoda and Hydracarina both exhibited positive correlations on *P. nodosus* and *N. odorata* and negative correlations on *H. verticillata*. The correlation for Copepoda on *P. nodosus* was significantly positive ($P \leq 0.001$) and moderately large ($r = 0.88$), whereas the correlation for Hydracarina on *P. nodosus* was weak ($r = 0.21$). The negative correlation between Hydracarina and *H. verticillata* surface area was strong ($r = -0.53$). Ostracoda abundance was negatively correlated to the surface area of *H. verticillata* ($r = -0.23$) and *N. odorata* ($r = -0.68$). However, Ostracoda abundance showed a significant positive correlation ($r = 0.74$) to surface area of *P. nodosus*.

Discussion

Krecker (1939) and Rosine (1955) have reported dissimilar densities from macrophyte species with different morphologies. Their findings indicate that macrophytes with highly dissected leaves support greater densities than plants with more simple leaf structure. Soszka (1975) and Gerking (1957) reported similar results. Beckett, Aartila, and Miller (in press) studied invertebrate colonization on *P. nodosus* in Eau Galle Lake in Wisconsin and reported the number of invertebrates per *P. nodosus* plant and per square centimeter of plant surface area. For June in Eau Galle lake, the mean invertebrate abundance was 154.9 individuals per mean surface area of 122.2 sq cm. For August, Beckett, Aartila, and Miller (in press) reported a mean of 127.4 invertebrates per mean surface area of 183.5 sq cm. Although Lake Seminole and Eau Galle Lake are geographically distant, both studies dealt with *P. nodosus*.

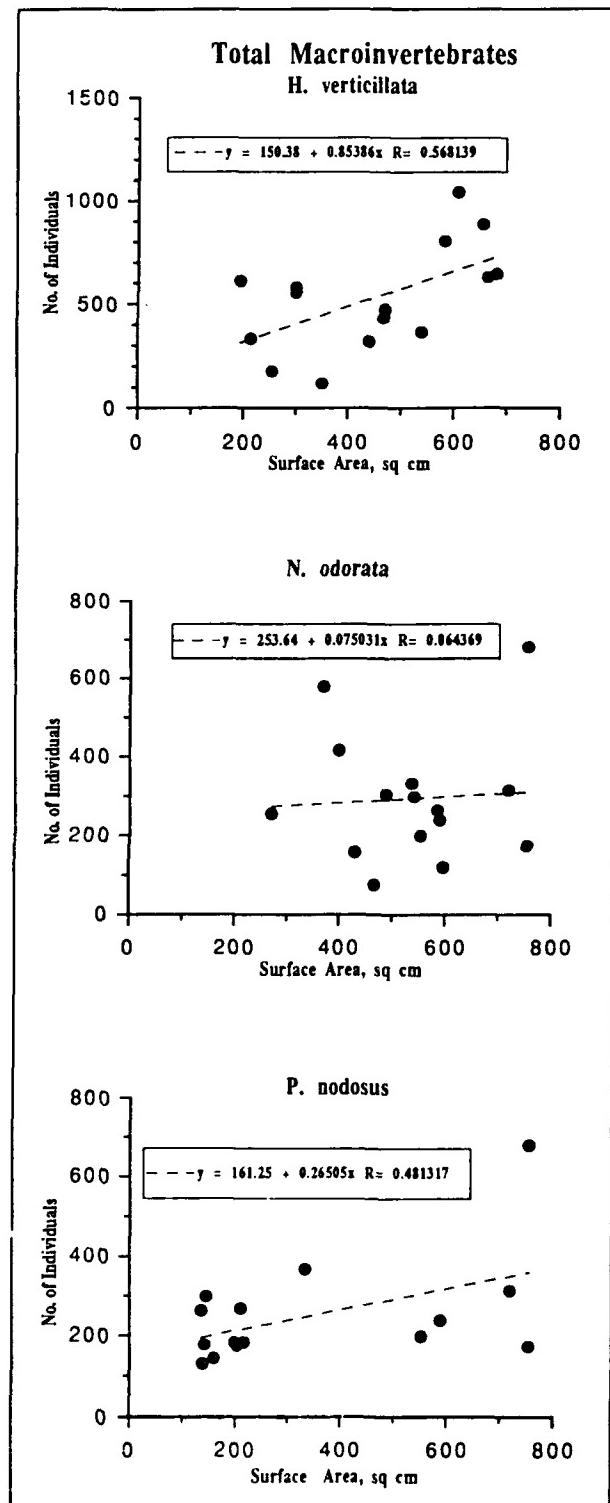


Figure 3. Relationship between total macroinvertebrates and surface area of three plant species

Total invertebrates per square meter were 12,676.4 in June 1987 for *P. nodosus* in Eau Galle Lake, whereas for Lake Seminole in July 1987, values were 9,689.0 invertebrates/sq m for *P. nodosus*, or approximately 1.3 times greater than the mean invertebrate density in Eau Galle Lake. Chironomid density in Lake Seminole was 1.8 times greater than chironomid density in Eau Galle Lake (6,187.1 individuals/sq m). Naidid density in Lake Seminole was approximately equal (1.1 times) to June naidid density values in Eau Galle Lake (2,012.3 individuals/sq m).

The growth pattern of macrophytes can influence invertebrate density. Since *H. verticillata* often branches repeatedly, a vast interconnecting macrophyte bed can be formed. Rooke (1986) noted that a complex growth pattern can lead to high invertebrate density, since invertebrates can find shelter from predators. The highest invertebrate mean (of the three plant species collected in Lake Seminole) was present on *H. verticillata*, a macrophyte that exhibits a complex growth pattern.

The chironomid assemblage on the three macrophyte species from Lake Seminole consisted of a total of 17 taxa, with three chironomid taxa dominating each of the three macrophyte species. Beckett, Aartila, and Miller (1992b) reported 21 chironomid taxa from *Ceratophyllum demersum*, a dominant macrophyte in Eau Galle Lake, Wisconsin, which is morphologically similar to *H. verticillata* (the dominant macrophyte species in Lake Seminole). Chironomidae on *P. nodosus* comprised over 56 percent of invertebrates collected, but chironomids were only 26.1 percent of total invertebrates collected on *H. verticillata*. Two chironomid taxa, *T. nr. fusca* and *Psectrocladius* sp., comprised over 79 percent of all chironomids collected on *P. nodosus*. Both of these taxa belong to Orthocladiinae, which feed by scraping algae from firm substrate (Merritt and Cummins 1978). The dominant chironomid on *H. verticillata*

was *Tanytarsus* sp. (a collector); however, this species comprised only 37.7 percent of the total chironomids collected on *H. verticillata*.

Rosine (1955) studied invertebrate densities on three macrophyte species in a Colorado lake. Rosine concluded that surface area was a determinant in invertebrate density and that plant species with dissected leaves would support higher densities than a plant species with nondissected leaves. Biochino and Biochino (1979) studied the relationship between invertebrate density and plant biomass and plant surface area. They concluded that plant surface area was significantly correlated with invertebrate density.

Correlations between surface area and invertebrate density revealed seven significant correlations. Five significant relationships existed between taxonomic groups on *P. nodosus*, and two between taxonomic groups on *H. verticillata*. Total invertebrate numbers were positively correlated with surface area of *H. verticillata*. *Hydrilla verticillata*, depending on stem length, can provide invertebrates with a large amount of colonizable substrate; it is not surprising that total invertebrate density was significantly related to surface area. The naidid abundance was significantly correlated to surface area of *H. verticillata*. Perhaps this correlation is due to the greater amount of detritus trapped by *H. verticillata*. Chironomidae were not correlated to surface area of *H. verticillata*, a macrophyte species with a large potential surface area. Since many of the chironomid taxa were collector/gatherers, these *P. nodosus* may not have had suitable epiphytic growth.

The presence of two leaf shapes and leaf placements (floating and submersed) may be important. *Nymphaea odorata* exhibited no significant correlations between invertebrate density and surface area. Low correlation may have been the result of lack of substrate heterogeneity. Since there were no places for organisms to hide from predators, a larger surface area did not benefit prey organisms.

Summary

In addition to macroinvertebrates, aquatic plants are important for immature and adult fishes, amphibians, and reptiles. By forming dense beds, macrophytes can serve as a nursery to fish by providing substrate for reproduction and a refuge from predation (Chilton 1990). Epiphytic invertebrates, such as those collected in this study, are important in the diet of fishes. In addition, waterfowl have been shown to feed on macrophyte tissue and epiphytic invertebrates (Krull 1970).

Problem levels of aquatic macrophytes can interfere with recreational and commercial uses of water bodies. However, an understanding of the value of aquatic plants for macroinvertebrates, as well as higher trophic-level organisms, can encourage the use of environmentally sound control strategies.

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Invertebrate Foods and Growth of Juvenile Largemouth Bass in Two Species of Aquatic Macrophytes

by

James V. Morrow,¹ Jan Jeffrey Hoover,¹ and K. Jack Killgore¹

Introduction

Largemouth bass populations are influenced by amount and spatial distribution of aquatic plants (Durocher, Procine, and Kraai 1984; Engel 1987; Colle, Cailteux, and Shireman 1989). Aquatic plants affect prey availability (Crowder and Cooper 1982), bass behavior (Savino and Stein 1982), feeding efficiency (Heck and Thoman 1981, Savino and Stein 1982), and selection of prey (Schramm and Zale 1985). Such plant-mediated impacts on feeding can ultimately influence condition (Colle and Shireman 1980), growth rates (Morrow, Killgore, and Hoover 1991), year class strength (Aggus and Elliott 1975), and production (Wiley et al. 1984) of largemouth bass. However, aquatic plants are highly variable in morphology, growth rate, and biomass. As a result, different plant species may exhibit substantial disparities in invertebrate abundance and community composition, thus influencing bass feeding behavior and growth.

In this study, we compared growth of juvenile largemouth bass in experimental ponds planted with similar densities and surface coverage of hydrilla (*Hydrilla verticillata*) and longleaf pondweed (*Potamogeton nodosus*). Hydrilla is an exotic species introduced from Asia (Tarver et al. 1978), whereas longleaf pondweed is a native aquatic plant occurring in many habitats and under a wide range of physical conditions (Beal 1977, Nelson and Couch 1985).

We tested the following null hypotheses: (a) community composition and relative abundance of invertebrates did not differ among

ponds and (b) growth of largemouth bass did not differ among ponds. Acceptance of these hypotheses would suggest that these two submerged plants provide similar aquatic habitat conditions for invertebrates and fish if plant density is comparable.

Methods and Materials

This study was conducted in six ponds at the Lewisville Aquatic Ecosystem Research Facility in Lewisville, TX. The dimensions of each pond were 28 by 70 m, with a maximum depth of 2 m near the outflow sloping to 0.5 m at the opposite end. The surface area of each pond was approximately 2,000 m². All ponds were treated with metam-sodium at 63 L/ha prior to filling to reduce occurrence of extraneous plant species.

During mid-May, three ponds (P-1, P-2, P-3) were planted with *P. nodosus*, and three ponds (H-1, H-2, H-3) were planted with *H. verticillata*. Pondweed tubers were planted about 45 cm apart in eight 10- by 10-m squares as the ponds were being filled. This created a checkerboard pattern of planted and unplanted areas with planted areas covering 800 m² or approximately 40 percent of the pond bottom. In the hydrilla ponds, short stems were planted in the same pattern as the pondweed ponds.

Juvenile largemouth bass were stocked into the ponds on 19 June, at which time stands of *P. nodosus* and *H. verticillata* were well established. The bass were acquired from a local hatchery and had been graded into groups of larger (65- to 90-mm) and

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smaller (40- to 65-mm) individuals and packaged in six bags of 100 of each grade.

Ponds were stocked in the following manner: one bag of large bass and one bag of small bass were released into a 132-L container and allowed to mix; 50 bass were removed and preserved as prestock sample to determine mean lengths; and the remaining 150 bass were released into the pond.

Invertebrate samples were collected with a D-shaped dip net having a 0.062-m² opening and 1-mm mesh. Five samples were taken from each pond at the time of largemouth bass stocking and at the time of recapture. Each sample consisted of one 15-m sweep from the center of the pond to the shore, perpendicular to the long axis of the pond. In the laboratory, invertebrates were identified to family or order and counted.

The nine invertebrate taxa that comprised approximately 90 percent of all individuals were used to evaluate differences among ponds. Differences in invertebrate assemblages among ponds were quantified using a measure of Euclidian distance, i.e., mean absolute difference (Ludwig and Reynolds 1988).

Mean absolute distance (MAD) is a function that evaluates overall resemblance among samples based on pairwise comparisons of numbers collected for a list of taxa. It differs from other similarity measures by considering absolute numbers collected (rather than percentages); it differs from other Euclidian measures by exhibiting greater sensitivity for differences in low-density taxa, and by standardization for taxonomic richness. Values can range from zero (i.e., numbers identical between ponds for all taxa) to infinity (i.e., no taxa common between ponds).

Just prior to draining ponds, plant biomass samples were taken by placing a 0.5- by 0.5-m polyvinyl chloride quadrant on the pond bottom and pulling up all plants within the square, excluding growth beneath the substrate. Samples were placed in 50- by 80-cm

mesh bags and spun by hand until no excess water spray was apparent. Wet weight was measured on a Pennsylvania I-10-L scale. Every other sample was saved, dried at 55 °C, and weighed on a Mettler PM16 scale.

A linear regression of wet weight to dry weight was determined using the General Linear Model procedure in SAS (Statistical Analysis System (SAS) 1985). The slope and intercept of this regression were used for wet weight/dry weight conversion of biomass.

Ponds H-1, H-2, and P-1 were drained on 9 October, and ponds H-3, P-1, and P-2 on 10 October. As the ponds were drained, all fish were collected and preserved. Largemouth bass were later measured for total length to the nearest millimeter. Total length values from prestock samples were similar in range and mean with no differences detected between ponds; therefore, length at recapture was used to determine growth. Differences in mean lengths were detected using the Least Significant Difference option of the GLM procedure in SAS (SAS 1985).

Stomachs were removed from 10 bass from each pond. Foods items were identified, counted, and percent volume estimated.

Results

Biomass and water quality

The surface of ponds H-1 and H-3 was completely covered with hydrilla at the end of the study. Ponds H-2, P-1, P-2, and P-3 retained the checkerboard pattern of the target species (40 to 46 percent surface coverage). However, all ponds had well-established stands of *Chara* sp. and *Najas* sp. in unplanted areas (Table 1).

Water temperatures ranged from 31 °C in early July to 25 °C in early October. Dissolved oxygen in all ponds was high, with saturation sometimes greater than 100 percent in the surface waters. Thermal and chemical stratification was evident in all ponds at

Table 1
**Wet-Weight Biomass of Aquatic Plants in Planted
 and Nonplanted Areas of Experimental Ponds¹**

Pond	Species Planted	Percent Coverage Target Species	Planted Areas		Nonplanted Areas	
			Wet-Weight Biomass, g/m ² (x 1000)	Standard Deviation (x 1000)	Wet-Weight Biomass, g/m ² (x 1000)	Standard Deviation (x 1000)
H-1	<i>H. verticillata</i>	100	15.9 ²	3.6	—	—
H-2	<i>H. verticillata</i>	40	10.1	5.3	8.1	7.9
H-3	<i>H. verticillata</i>	100	12.0	2.4	—	—
P-1	<i>P. nodosus</i>	44	8.1	2.4	9.0	3.1
P-2	<i>P. nodosus</i>	46	11.2	2.8	10.4	2.8
P-3	<i>P. nodosus</i>	45	11.5	4.0	9.6	9.1

¹ Biomass in nonplanted areas consisted of *Najas* sp., *Chara*, and *H. verticillata*.

² Significantly greater than H-2, P-1, and P-3 ($P = 0.05$).

different times but was not consistent, indicating that each pond mixed several times during the study. Water was neutral ($\text{pH} = 7$) to alkaline ($\text{pH} = 12$).

Food resources

Invertebrate assemblages were comparable among ponds, including those with different densities and species of plants, but temporal changes in invertebrate density were pronounced. Values for mean absolute distance of invertebrate assemblages on either date were no higher between hydrilla and pondweed ponds than among ponds of hydrilla or pondweed ($\times \text{MAD} 21.0 - 29.4$); however, values were substantially higher when comparing assemblages at the start of the experiment with those at the end of the experiment, for hydrilla ponds ($\times \text{MAD} = 58.6$) and for the pondweed ponds ($\times \text{MAD} = 75.9$).

Initially, invertebrate densities were low (most <200 individuals/sample), dominated by limnetic and climbing forms: Cladocera (water fleas), Notonectidae (back swimmers), and Coenagrionidae (narrow-winged damselflies) (Figure 1). At the conclusion of the experiment, densities were higher ($>350/\text{sample}$), especially in hydrilla ponds ($>450/\text{sample}$), and assemblages were dominated by benthic and sprawling forms: Gastropoda (snails), Libellulidae (common skimmers), and Chironomidae (larval midges) (Figure 2). Libellulidae was the most common food in

numbers (and volume) for bass from all ponds (Figures 3 and 4) and was the only taxon found in bass stomachs from all ponds.

Growth

Total length values of largemouth bass at stocking were the same between ponds (mean 60 to 63 mm; coefficient of variation 18 to 20 percent). Therefore, total length at recapture was used as an indicator of growth. Bass from H-2 had the highest growth, with a mean length of 214 mm at the end of the study (Table 2). Growth in H-2 was significantly greater than in all other ponds ($P \leq 0.05$). The lowest growth of bass was observed in H-1 (192 mm at the end of the study), significantly lower than ponds H-2, P-1, and P-3.

The mean length of bass in the hydrilla ponds was greater than in the pondweed ponds at the end of the study. The difference was small (7 mm) but statistically significant ($P < 0.05$). Growth of bass was not positively associated with total numbers of invertebrates, numbers of an individual taxa, or plant biomass.

Discussion

Mean length of juvenile largemouth bass in this study exceeded the national value, 116 mm (Carlander 1977), and the young of the year mean length for Florida largemouth bass, 175 mm (Coleman 1984). These high

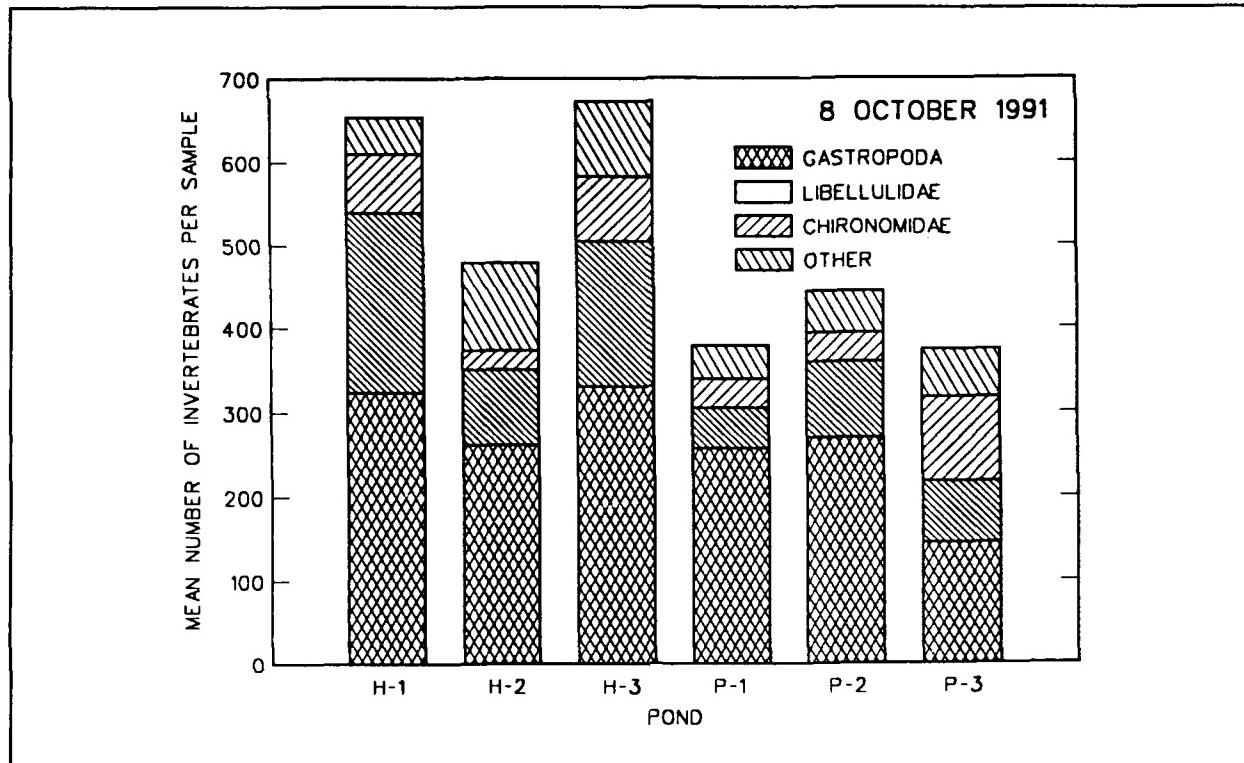


Figure 1. Mean number of invertebrates in experimental ponds before stocking largemouth bass

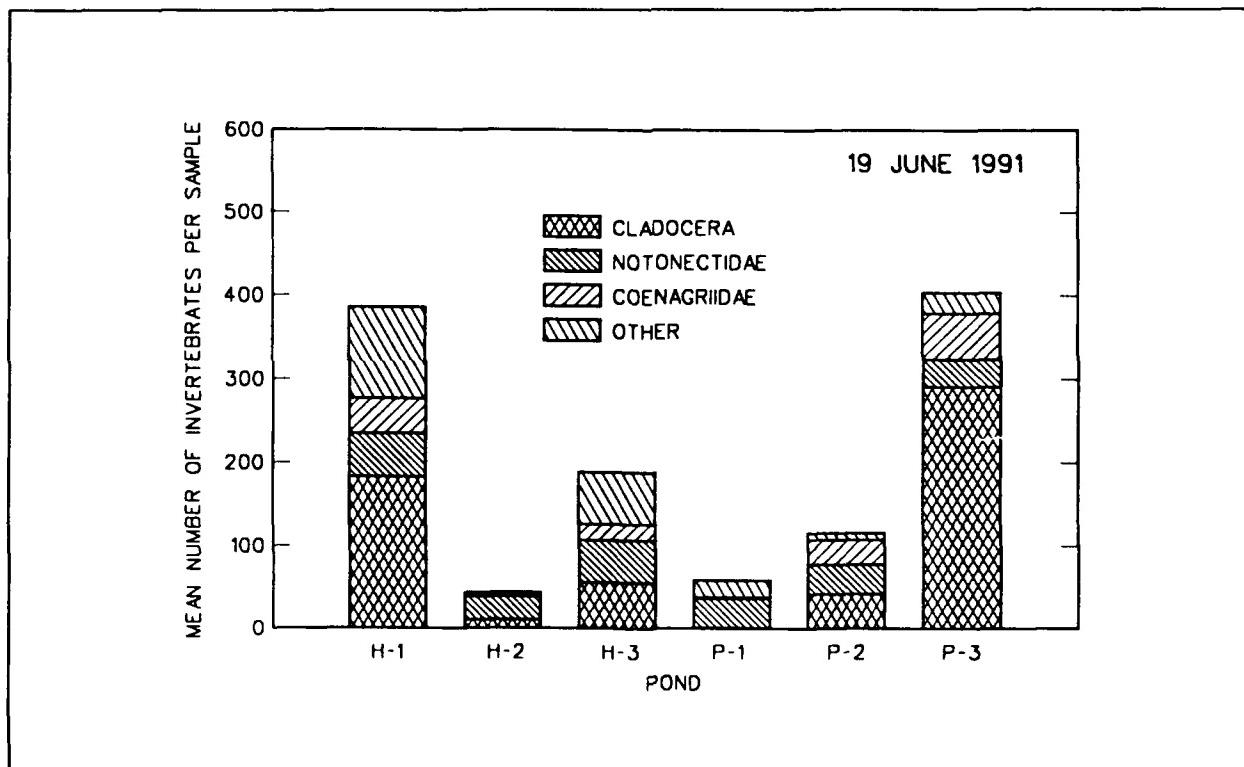


Figure 2. Mean number of invertebrates in experimental ponds immediately before harvesting largemouth bass at the end of the study

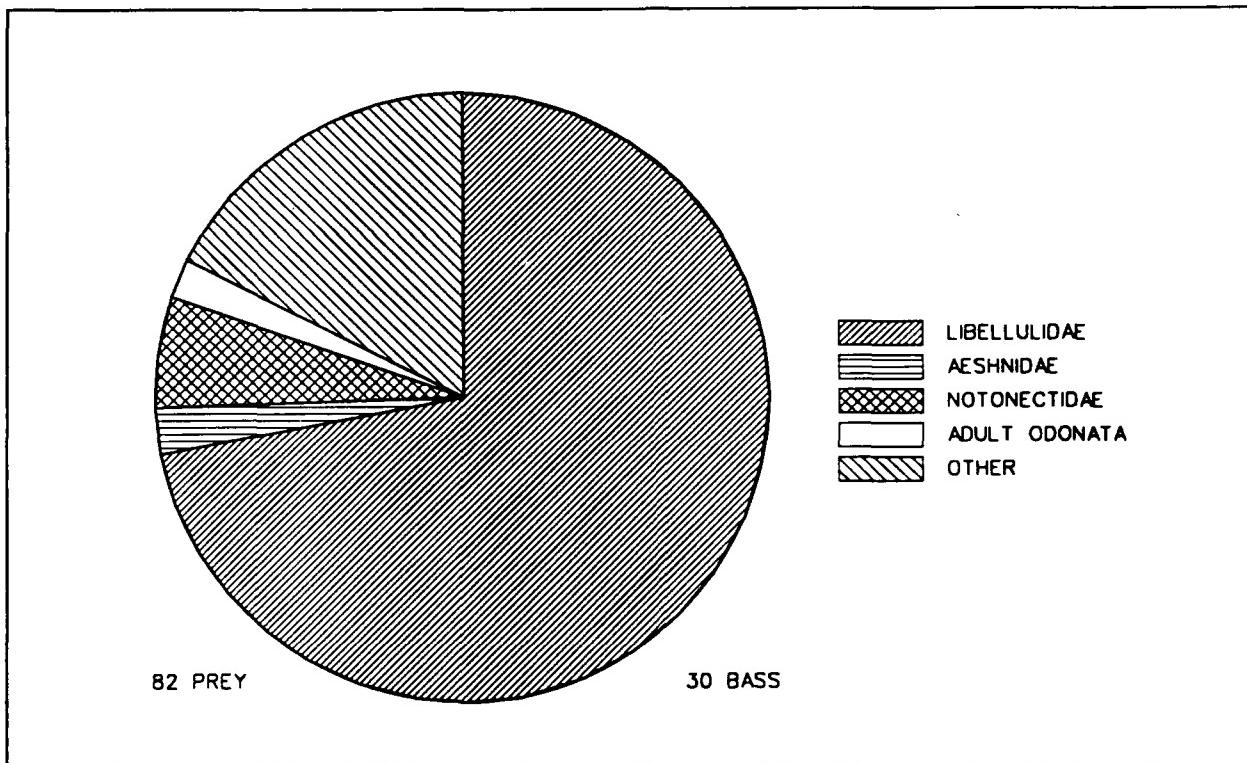


Figure 3. Composition of food items in largemouth bass stomachs from the *hydrilla* ponds

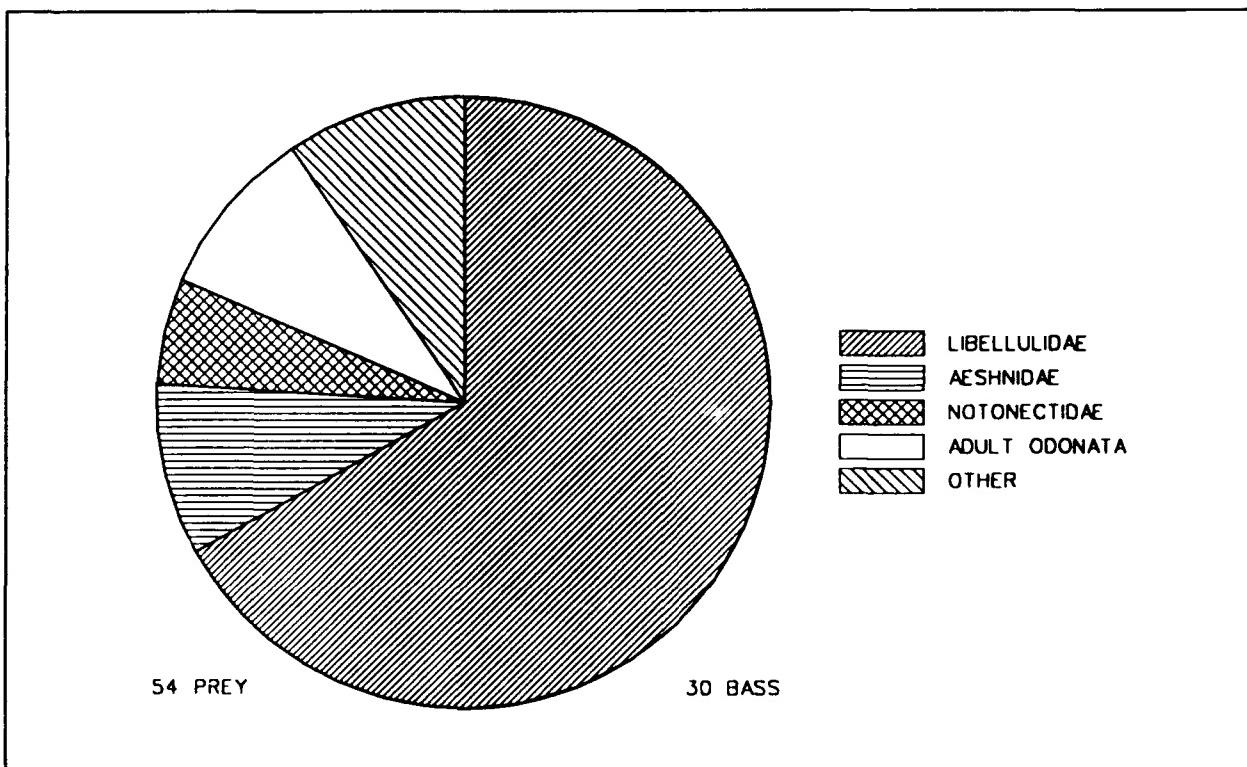


Figure 4. Composition of food items in largemouth bass stomachs from the pondweed ponds

Table 2
Mean Total Length of Largemouth Bass
at Time of Harvest

Pond	Species Planted	N	Mean Total Length, mm	Standard Deviation
H-1	<i>H. verticillata</i>	111	192 ¹	10.5
H-2	<i>H. verticillata</i>	138	214 ²	15.0
H-3	<i>H. verticillata</i>	117	195	11.9
P-1	<i>P. nodosus</i>	115	199	11.8
P-2	<i>P. nodosus</i>	117	194	11.1
P-3	<i>P. nodosus</i>	116	198	13.1

¹ Significantly less than total lengths from H-2, P-1, and P-3 ($P = 0.05$).

² Significantly greater than all other lengths ($P = 0.05$).

growth rates were achieved in spite of a high stocking rate of 750 bass per hectare and an absence of forage fish. However, water temperatures were near optimum for largemouth bass growth (27 °C. Coutant 1975) throughout the study, and high plant production and pH suggest that the ponds were fertile.

The large numbers of largemouth bass recovered from the ponds (Table 2) indicate that a high rate of cannibalism did not occur and that the bass achieved most of their growth on a diet of invertebrates. Substantial differences were not found between invertebrate communities in pondweed ponds and hydrilla ponds at the start of the study. However, differences were evident at the end of the study, with hydrilla having higher densities than pondweed.

Abundance of phytophilous invertebrates has been shown to depend more on surface area of aquatic plants than their weight (Biochino and Biochino 1979). Hydrilla has long branching stems, with small leaves 4 mm wide and 8 to 18 mm long, occurring in whorls of three to five (Tarver et al. 1978). This type of growth provides a substantial surface area per stem length, resulting in more macroinvertebrates inhabiting hydrilla than pondweed (Peets 1991). Although invertebrate colonization is slightly higher on hydrilla, no positive relationship between numbers of invertebrates and growth of largemouth bass was evident.

Growth of bass was significantly greater, by 7 mm or 3.5 percent, in hydrilla ponds than in pondweed ponds, but the overall difference of 7 mm was attributable to the exceptional growth in pond H-2; bass in the other hydrilla ponds grew at the same rate or less than those in pondweed ponds. Since the plant surface coverage of H-2 was 40 percent compared to 100 percent in H-1 and H-3, higher growth of juvenile bass may be related to spatial distribution of the plants. Therefore, this study indicates that similar densities and spatial patterns of hydrilla and longleaf pondweed provide comparable habitat for juvenile largemouth bass. When plant surface coverage is different, however, juvenile bass may grow faster in areas of intermediate coverage.

Growth of largemouth bass in vegetated habitats is directly related to plant density and spatial configuration of the plant bed. Colle and Shireman (1980) indicated that condition of small largemouth bass was lower when hydrilla coverage exceeded 50 percent than at lower levels. At low plant densities, largemouth bass actively searched for prey, switching to ambush techniques in higher plant densities, with feeding success impaired at very high densities, i.e. >250 stems/m² (Savino and Stein 1982). Engel (1987) demonstrated that bass over 180 mm rarely penetrated continuous plant beds but would use lanes cut into the beds.

Optimum habitat for largemouth bass in vegetated water bodies includes dense plant beds interspersed by open water. However, this study suggests that growth of largemouth bass during their first growing season can be rapid in areas of extremely high densities of plants of morphologically different species. An abundant food supply and cover from predators provide conditions to enhance growth and survival of juvenile bass. As bass continue to grow, they move into deeper water and associate with edges of the plant bed. Therefore, creating edges (by mechanical harvesting or herbicides) for use by older bass and preserving areas of dense plant beds (for use as nursery areas) are recommended.

if fishery management is an objective of the aquatic plant control program.

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Effects of Nighttime Convective Circulation on Phosphorus Dynamics Between Littoral and Pelagic Zones in Eau Galle Reservoir, Wisconsin

by

William F. James¹ and John W. Barko¹

Introduction

Most investigations of internal phosphorus (P) loading in lakes have concentrated on P release from profundal sediments under anaerobic conditions (e.g. Riley and Repas 1984, Nürnberg 1987). However, recent studies have shown that P can also be released from littoral sediments under aerobic conditions (Twinch and Peters 1984, Drake and Heaney 1987). Phosphorus released during the senescence of aquatic macrophytes constitutes another potential source of internal P loading from the littoral zone (Barko and Smart 1980, Carpenter 1980). Since internal P loading from littoral sources may significantly influence the P economy of aquatic systems, particularly small lakes (Prentki et al. 1979), it is important to evaluate horizontal P exchanges between littoral and pelagic zones of these systems (James and Barko 1991).

We report the results of seasonal investigations of P exchange between the littoral and pelagic zones of Eau Galle Reservoir, Wisconsin, driven by nighttime convective circulation. Because of the importance of P gradients in affecting P flux rates during these periods of circulation, we examine profiles of P in the overlying water column. Rates of P release from littoral sediments, as a function of both pH and oxygen environment, are estimated from results of laboratory incubation studies employing intact sediment cores. The objective of this article is to document the importance of convective circulation as a means of transporting P, as well as other constituents

such as herbicides, from the littoral to the pelagic zone.

Methods

Rates of phosphorus release from littoral sediments

Rates of soluble reactive P (SRP) release from littoral sediments were measured in the laboratory at pH values ranging from 8.0 to 10.0 under both aerobic and anaerobic conditions. Intact sediment cores collected from the littoral zone were incubated with overlying water at 20 °C for 1 to 2 weeks in core liners sealed with rubber stoppers. Rates of SRP release from littoral sediments ($\text{mg m}^{-2} \text{ day}^{-1}$) were calculated as the change in SRP in the overlying water divided by time and the cross-sectional area of the sediment core liner.

Limnological profiling

Six stations were established along a transect located in the northwest bay region of the reservoir (Figure 1) for monitoring seasonal changes in limnological conditions pertinent to the investigation. Vertical profiles of pH, dissolved oxygen (DO), total P (TP), and SRP were obtained at these stations over biweekly intervals from June through early September 1989. Conditions of pH and DO were measured with a Hydrolab Surveyor II, which was precalibrated with buffers and Winkler DO determinations (American Public Health Association (APHA) 1985).

¹ US Army Engineer Waterways Experiment Station, Vicksburg, MS.

Samples for both TP and SRP analyses were collected with pneumatically driven close-interval syringe samplers, as described in James and Barko (1991). TP was analyzed on a Technicon Auto-Analyzer II following digestion with potassium persulfate (APHA 1985). SRP was analyzed colorimetrically on a Perkin-Elmer Lambda 3b spectrophotometer using the ascorbic acid method (APHA 1985).

Exchanges between littoral and pelagic zones

Hydraulic exchange rates during nighttime cooling were determined from patterns of dye dispersion between the littoral and pelagic zones. A series of transects were established in the vegetated region of the northwest bay,

along the slope of the basin and perpendicular to it (Figure 1). Hydraulic exchange rates were determined at fixed biweekly intervals from June through August 1989 using Rhodamine WT red fluorescent dye (Crompton and Knowles, Inc.) as a tracer.

Methods for determining hydraulic exchange rates from dye dispersion were similar to those described in James and Barko (1991). Water temperature was measured every half hour at stations 1, 2, 5, and 6 (Figure 1) during each dye study period using recording thermistor strings (OmniData International) calibrated to the nearest 0.1 °C.

TP exchange rates ($\text{mg m}^{-2} \text{ hr}^{-1}$) into the littoral or pelagic zone were calculated as the product of hourly volumetric flow rates and depth-integrated TP in the region of flow divided by the reservoir surface area. Net TP flux rates ($\text{mg m}^{-2} \text{ hr}^{-1}$) were calculated as the difference between TP exchange rates into littoral and pelagic zones.

Results

Littoral-pelagic P gradients

Rates of SRP release from littoral sediments, measured in the laboratory at 20 °C, increased with increasing pH under both aerobic and anaerobic conditions (Figure 2). At lower values of pH, sediments incubated under anaerobic conditions exhibited higher rates of SRP release than under aerobic conditions. However, near pH 10, sediments exhibited similar rates of SRP release under aerobic and anaerobic conditions. These data were used to establish rates of SRP release from littoral sediments in the reservoir, based on pH and DO measured in the reservoir.

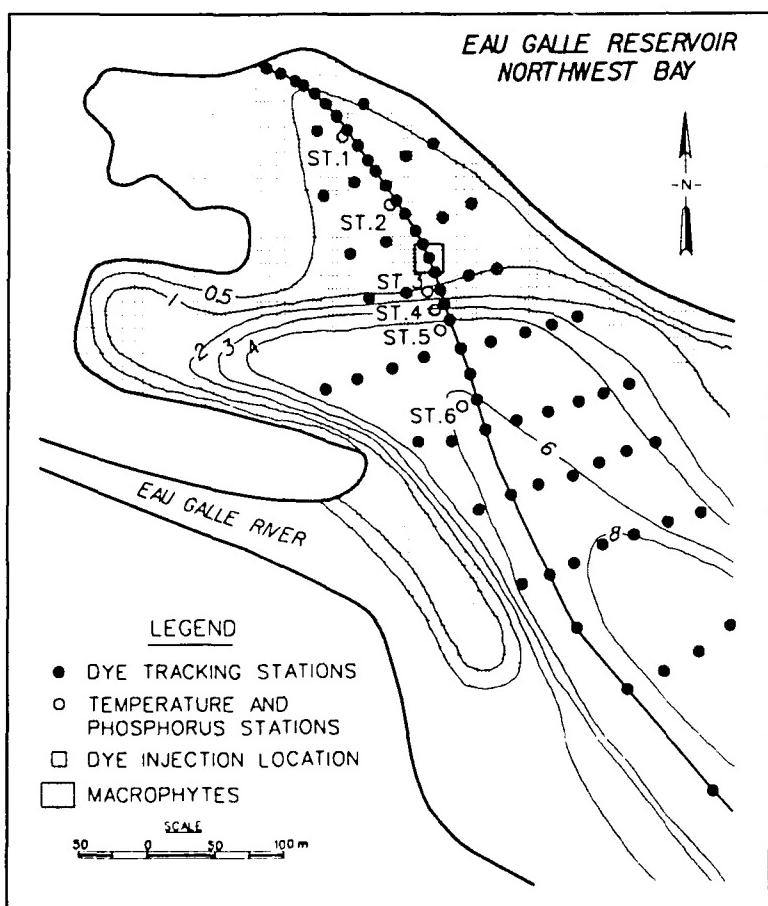


Figure 1. Morphometric contours (meters) and station locations in northwest bay region of Eau Galle Reservoir. Shaded area represents regions of macrophyte growth (littoral zone)

Marked horizontal and vertical gradients in TP developed in the littoral zone from May through August (Figure 3), and SRP accounted for up to 45 to 90 percent of the measured TP. TP was generally much lower in the epilimnion of the pelagic zone than in the littoral zone.

Convective circulation

During each dye tracking investigation, the littoral zone exhibited cooler depth-integrated water temperatures than the pelagic epilimnion during the night and morning hours. These differences in water temperature resulted in convective circulation. Dye dispersed as a bottom current from the littoral zone toward the pelagic zone at night (Figure 4). Intrusion of littoral water into the pelagic zone occurred as an interflow, consistently confined to 0.25 m in vertical expanse.

The temperature of this interflow coincided with temperatures measured in the littoral zone (Figure 4). As the result of seasonal variations in pelagic thermal stratification, the depth of interflow decreased to a minimum in July, then increased in August.

P exchange dynamics

Hourly TP exchange rates to both the littoral and pelagic zones were greatest in early July and early August, and lowest in June (Table 1). Estimated hourly TP exchange rates to the pelagic zone were greater than to the littoral zone, due to higher TP in the littoral bottom waters. Net TP flux to the pelagic zone, at rates ranging between 0.12 and 0.43 mg m⁻² hr⁻¹, occurred during all dye tracking investigations (Table 1). A mean net TP flux of 0.22 mg m⁻² hr⁻¹ to the pelagic zone during nighttime circulation was estimated for the midsummer period.

Discussion

Rates of P release from littoral sediments of Eau Galle Reservoir, measured in the labo-

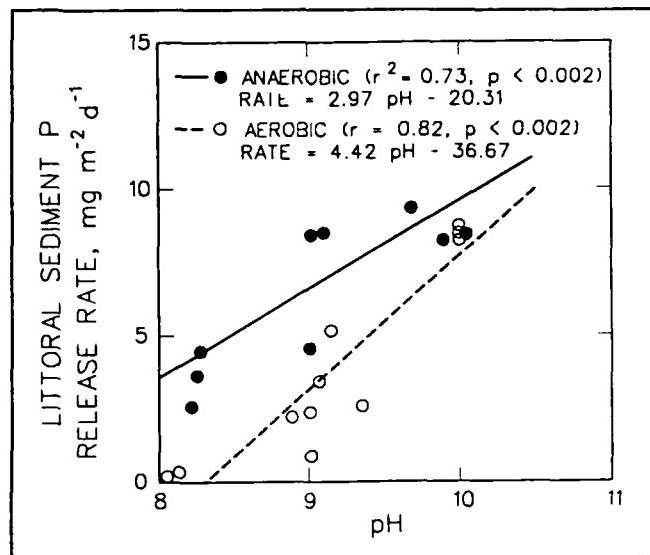


Figure 2. Variations in rates of SRP release from littoral sediments as a function of pH under aerobic and anaerobic conditions for undisturbed laboratory sediment incubation systems

ratory, were linearly related to pH under both aerobic and anaerobic conditions. As in our investigation, other studies have shown that pH can have a regulating effect on rates of P release from sediments, particularly under aerobic conditions (Lee, Sonzogni, and Spear 1977; Drake and Heaney 1987).

TP gradients in the littoral zone were prominent throughout much of the summer, and SRP accounted for a large percentage of littoral TP. To our knowledge, no investigations have shown such marked concentration gradients vertically in the littoral zone or horizontally between littoral and pelagic zones. These observations, in combination with results obtained in our laboratory incubation systems, strongly suggest that littoral sediments provide an important source of P to the reservoir.

Nighttime convective circulation constituted an important mechanism for movement of littoral P to the pelagic zone during the summer. During the night and morning, cooler bottom water from the littoral zone moved as an interflow current into the pelagic zone at the base of the epilimnion, and

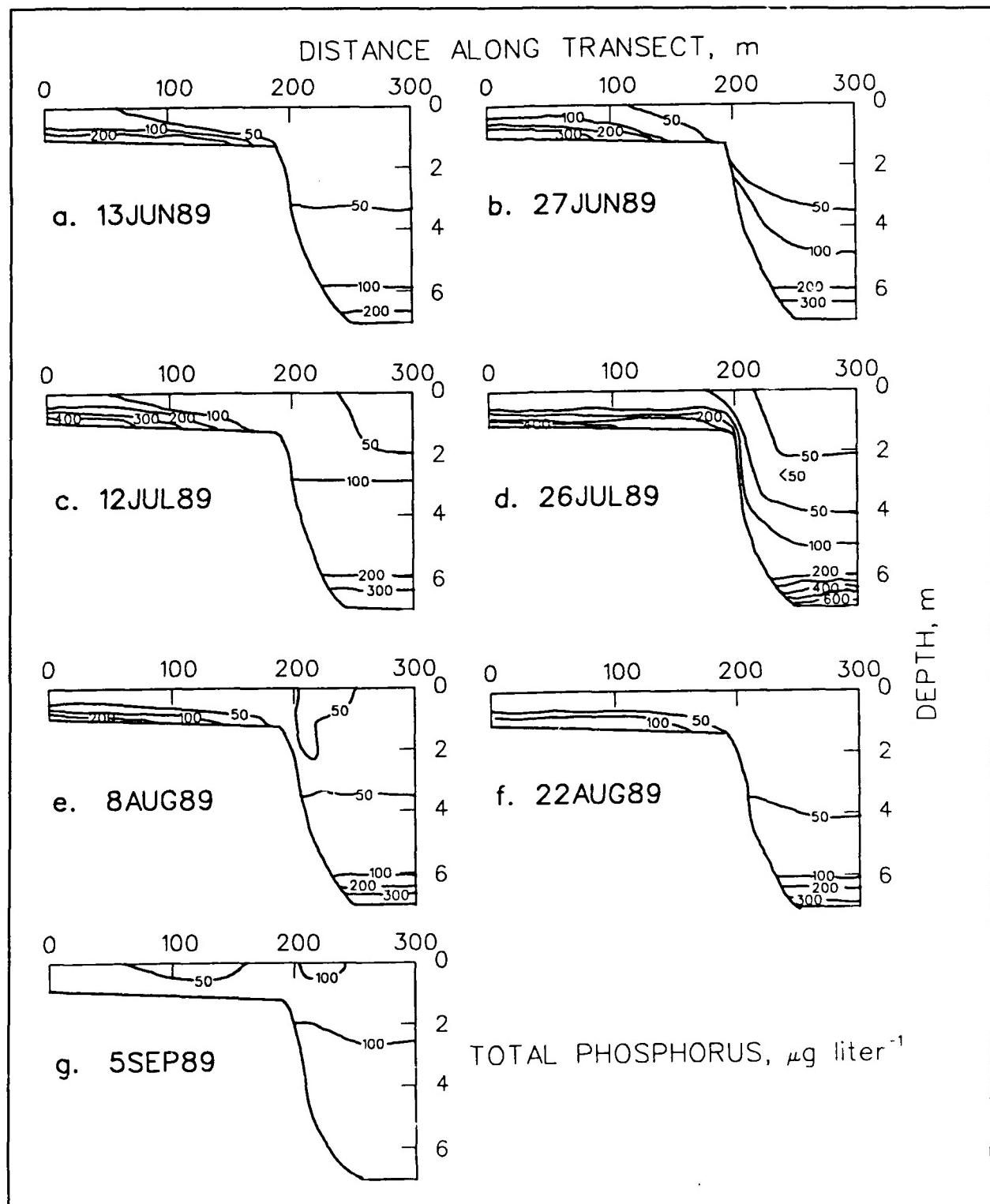
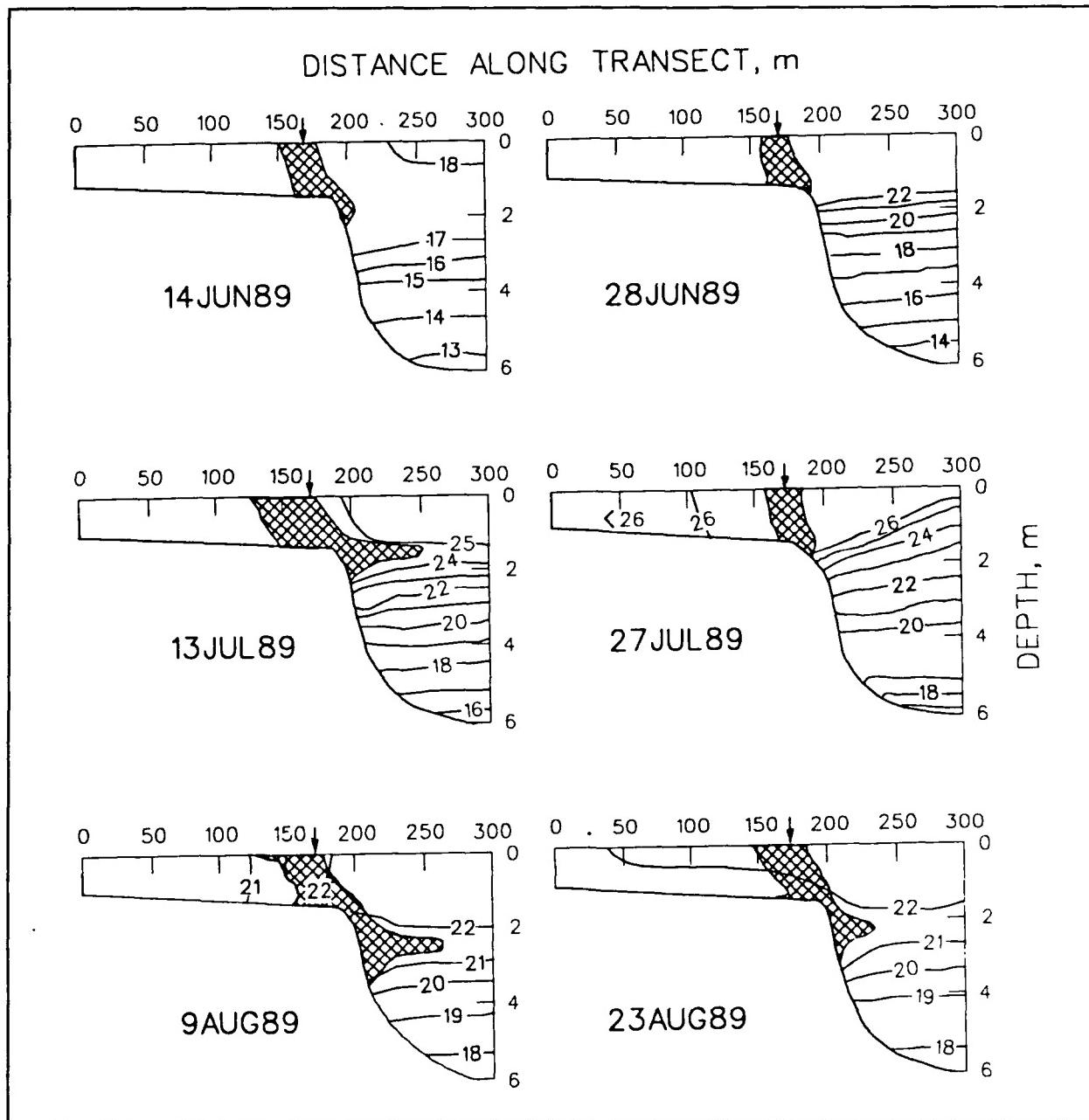


Figure 3. Seasonal, vertical, and horizontal variations in TP concentration in littoral and pelagic zones



plant management in this and other reservoirs. For instance, convective circulation may affect the residence time of aquatic herbicides and result in their transport to undesirable locations. In this regard, we have expanded our studies on convective circulation to Guntersville Reservoir, Alabama, which has experienced aquatic macrophyte problems. In cooperation with the Tennessee Valley Authority, we are currently examining this hydraulic transport mechanism in relation to aquatic plant management through herbicide application.

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Table 1
**Estimation of Hourly TP Exchange Rates
Between Littoral and Pelagic Zones, and Net TP Flux
to Pelagic Zone During Dye Tracking Periods**

Date	Direction of Movement	TP mg m ⁻³	Hourly TP Exchange Rate mg m ⁻² hr ⁻¹	Net TP Flux ¹ mg m ⁻² hr ⁻¹
13-14 Jun 89	Pelagic Littoral	91.5 40.3	0.22 0.10	0.12
27-28 Jun 89	Pelagic Littoral	124.8 35.8	0.22 0.06	0.18
12-13 Jul 89	Pelagic Littoral	134.6 52.8	0.71 0.28	0.43
26-27 Jul 89	Pelagic Littoral	184.3 43.8	0.27 0.06	0.21
8-9 Aug 89	Pelagic Littoral	87.0 52.2	0.67 0.40	0.27
22-23 Aug 89	Pelagic Littoral	85.6 42.0	0.30 0.15	0.15

¹ Net TP flux to the pelagic zone is the difference between hourly TP exchange to the littoral and pelagic zones divided by the reservoir surface area (0.6 km²).

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Effects of Benthic Barrier Placements on Aquatic Habitat Conditions

by
Harry L. Eakin¹

Introduction

Among lake management strategies used to control or eradicate nuisance growths of rooted macrophytes, various techniques of covering the bottom of aquatic habitats have been employed since the late 1960s (Born et al. 1973, Nichols 1974). Successful application of this strategy is dependent on the ability of bottom covers to isolate the sediment as a substrate for plant utilization, and also to limit the plants' access to the overlying water column and sunlight. Techniques have ranged from overlaying the sediment surface with sand and gravel to placement of different types of sheeting, usually plastics or synthetic fabrics.

Over the past two decades, numerous investigations (e.g. Mayer 1978; Cooke and Gorman 1980; Lewis, Wile, and Painter 1983; Engel 1984) have examined the practicability and efficacy of various benthic barriers. While the positive and negative attributes of barrier placements are generally understood (Cooke 1986), previous research has provided little insight into the effects of benthic barriers on the environment, specifically the physical and chemical composition of sediments they contact.

The objectives of this study were to examine the effects of barrier placements on selected physical and chemical components of sediment. The investigation was conducted within the littoral regions of Lake Guntersville, Alabama, and Eau Galle Reservoir, Wisconsin, during 1990 and 1991.

Materials and Methods

Study sites

Lake Guntersville is a large (about 274-km²) Tennessee Valley Authority reservoir located in Jackson and Marshall Counties, Alabama, and Marion County, Tennessee. Benthic barriers were deployed on May 22, 1990, at five locations within the Town Creek embayment of Lake Guntersville (Figure 1). However, the benthic barrier at site 1 was subsequently vandalized and destroyed. Locations of barrier placements were chosen based on historical information² indicating past dominance at selected sites by *Hydrilla*. However, soon after barrier placement, a dramatic and almost total decline of submersed macrophytes within the study area occurred and persisted throughout the study period.

Eau Galle Reservoir is a small (0.62-km²) US Army Corps of Engineers impoundment located in west-central Wisconsin. Along the north shore of the reservoir, near the mouth of Lousy Creek, a single benthic barrier was deployed during late August 1989 in a plant bed containing *Potamogeton* but dominated by *Ceratophyllum* (Figure 2).

Sampling/analytical protocols

Three techniques were employed to examine the effects of barrier placement. Sediment cores were collected from beneath and adjacent to each barrier for comparison of physical and chemical parameters. In situ interstitial water samplers were deployed for assessment

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² Personal Communication, April 1990, Earl Burns and David Webb, Aquatic Biology Department, Tennessee Valley Authority, Muscle Shoals, AL.

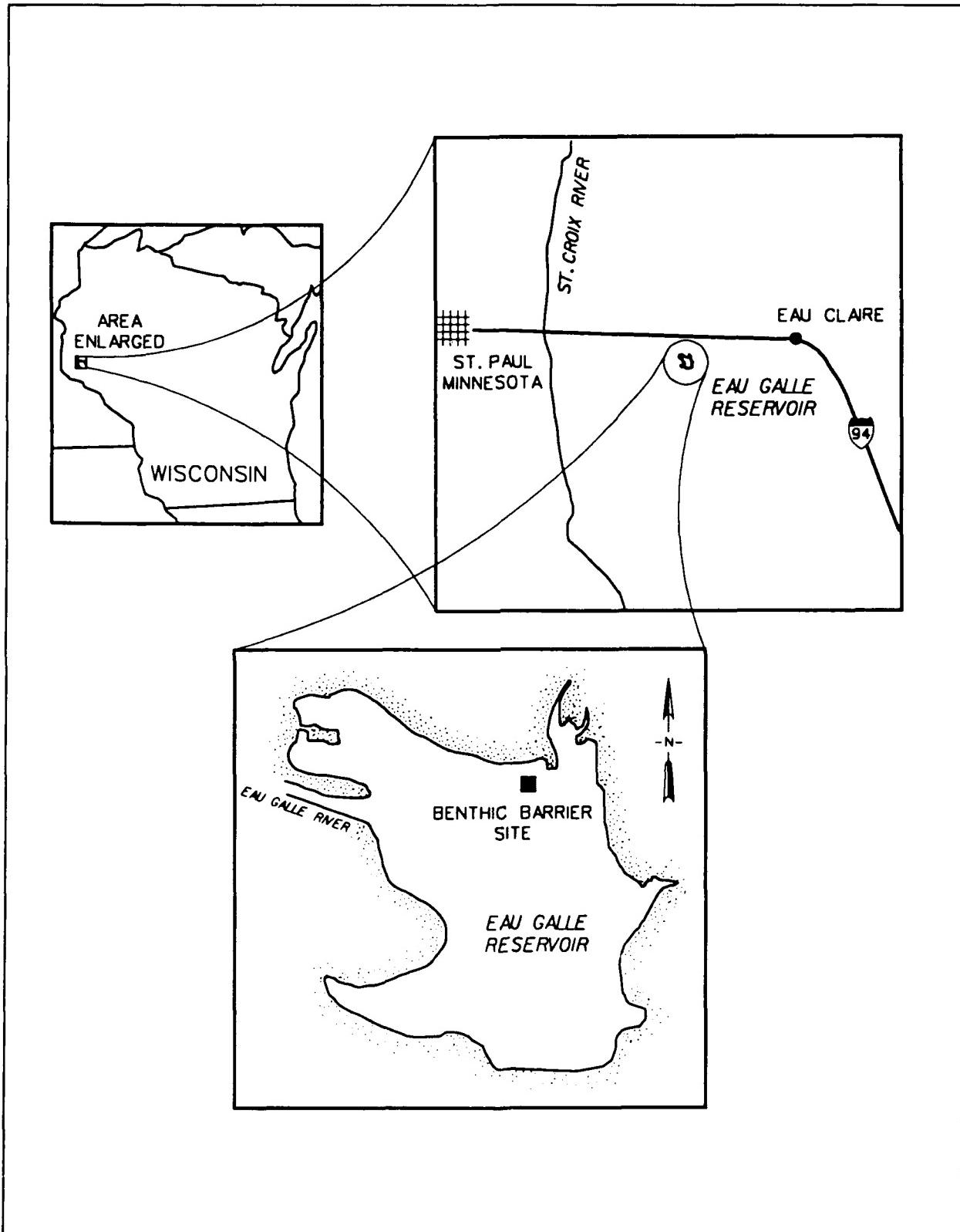


Figure 2. Benthic barrier sampling location in Eau Galle Reservoir

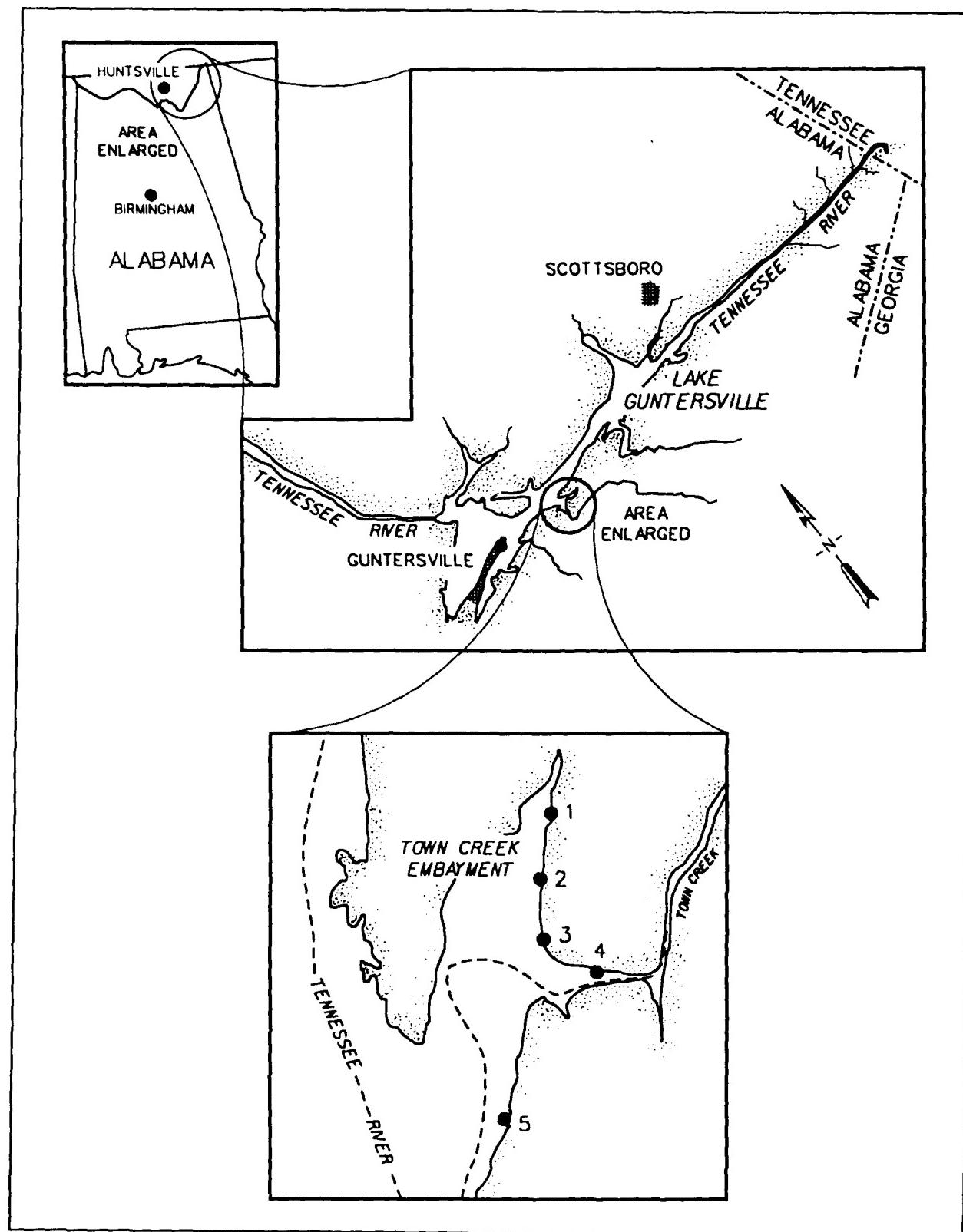


Figure 1. Benthic barrier sampling locations in Lake Guntersville

of soluble nutrient levels. In addition, dialysis chambers were placed beneath each barrier to measure dissolved oxygen (DO) concentrations in the sediment surficial water, i.e., water between the barrier and the sediment surface.

Sediment cores

Core samples were collected during June and September 1991 at Eau Galle Reservoir and during May and October 1991 at Lake Guntersville. Sampling times were selected to assess any physical or chemical changes occurring over the macrophyte growth season in each reservoir. Sample collection was performed with a Wildco hand core sampler (Wildlife Supply Company) equipped with acrylic core liners (6.5 cm inside diameter, 50 cm long). Only the top 10 cm of each core sample was retained for analysis. All core samples were refrigerated at 4 °C and transported to the laboratory for processing within 48 hr.

Under a nitrogen atmosphere within a glove box, each core sample was homogenized and subsampled for physical and chemical analysis. Moisture content, density, and organic matter content were measured by drying a known volume to a constant weight at 105 °C, then combusting at 550 °C (Allen et al. 1974). Following extraction with 1 M NaCl (Bremner 1965), exchangeable NH₄-N concentrations were determined colorimetrically on a Technicon AutoAnalyzer II using the salicylate-hypochlorite method (Technicon Corporation 1978). Likewise, extractable PO₄-P was measured colorimetrically using the ascorbic acid reduction method (American Public Health Association (APHA) 1985) after dilute acid extraction (Olsen and Sommers 1982). Total nitrogen and total phosphorus concentrations were also measured colorimetrically after digestion with H₂SO₄, K₂SO₄, and red HgO (Plumb 1981). Particle size determinations were made using a modification of a hydrometer method (Patrick 1958) first described by Day (1956).

Interstitial water

Plexiglas samplers, after a design of Hesslein (1976), were used to collect *in situ* close-interval interstitial (pore water) samples. The principle of operation of the samplers is based on the equilibration of a contained quantity of water with the surrounding water through a dialysis membrane (1.0-μm, polycarbonate, Nucleopore Corporation).

During a 2-week period in July and August 1990, a pilot experiment was conducted at site 4 in Lake Guntersville to establish an appropriate method to determine the effects of barrier placements on sediment pore water, for subsequent sampler deployments at Lake Guntersville and Eau Galle Reservoir. Replicate samplers were placed near the center, along the inside edge of the barrier, and in the open sediment adjacent to the barrier. Relying on the results from this pilot experiment, *in situ* pore water samples were collected in September 1990 and once per month during the 1991 growth season (June-September) beneath the center of each barrier.

Samplers were prepared using the techniques of Carignan (1984), to minimize oxidative precipitation of reduced species, and deployed vertically in sediment beneath the center of each barrier and in open sediment adjacent to each barrier. Samplers remained in place for about 14 days, a sufficient time period to allow equilibration (Carignan 1984). Upon retrieval, the contents of each close-interval chamber were removed, filtered (0.45 μm, Nalgene CA syringe filters), and preserved with H₂SO₄ to pH <2.

Interstitial water samples were analyzed for NH₄-N and soluble reactive PO₄-P, using the methods described above, and for Fe and Mn by direct atomic absorption spectrophotometry (APHA 1985).

Surficial water

Plexiglas dialysis chambers (L, 10.2 cm; W, 7.6 cm; H, 5.1 cm), containing about 400 ml of deoxygenated and deionized distilled water, were placed on the sediment surface beneath each barrier. Deployments of the surficial water samplers were coincident with in situ close-interval sampler deployments. Upon retrieval, the contained water was transferred to 300-ml biological oxygen demand bottles and "fixed" for later determination of DO concentrations by the azide modified iodometric method (APHA 1985).

Results and Discussion

Sediment cores

The physical and chemical composition of sediment examined at study sites in Lake Guntersville exhibited four distinct patterns (cf. Figure 3, Table 1). One pattern showed only minor differences in sediment type between sites 2-4, as well as between sampling locations (i.e., beneath or adjacent to the barrier placements) at these sites. A second pattern demonstrated considerable differences in sediment type between site 5 and sites 2-4. A third pattern showed, in many cases, substantial compositional differences between sediment collected beneath and adjacent to the barrier at site 5. And, a fourth pattern indicated no appreciable changes in sediment composition over time, as evidenced between the May and October 1991 sampling efforts at all sites.

Moisture content was about 22 percent (Figure 3) at sites 2-4 for both sampling periods and locations. Organic matter content at these sites was about 2.6 percent for both sampling periods and locations. At site 5, moisture content was about 48 and 62 percent beneath the barrier and open locations, respectively, for the May 7, 1991, sampling period. Organic matter content beneath the barrier at site 5 was 6.1 percent in comparison with 9.2 percent at the open location.

Decreased moisture content in sediment beneath the barrier may be attributable to degradation of organic matter, and the concomitant loss of water-holding capacity, over the period of barrier placement. On October 10, 1991, moisture content at site 5 was about 40 percent beneath the barrier and 53 percent at the open location. Similar reductions in moisture content of about 9 percent, both beneath the barrier and at the open location (site 5), appear to reflect consolidation processes.

Exchangeable NH₄-N concentrations observed at sites 2-4 were similar for both sampling periods and locations. At site 5, however, NH₄-N concentrations were clearly different (both beneath the barrier and at the open location) than at respective locations at sites 2-4. Higher NH₄-N levels measured beneath the barrier at site 5 may have resulted from greater microbial activity associated with higher organic matter content.

Total nitrogen concentrations at sites 2-4 were comparable for both sampling periods and locations. However, in contrast to exchangeable NH₄-N concentrations at site 5, total nitrogen levels were lower beneath the barrier and higher in the open sediment for both sampling periods. Comparison of total nitrogen levels at site 5 suggests greater overall decomposition of organic matter in sediment beneath the barrier, since the time of barrier placement, than at the open location.

Barko and Smart (1986) found significant positive correlations between total nitrogen levels and organic matter content within sediments. Concurrent decreases in total nitrogen, moisture content, and organic matter levels (Table 1) indicated similar rates of decomposition of organic matter in sediments beneath and adjacent to the barrier over the 1991 plant growing season.

Patterns observed in sediment composition at Lake Guntersville tend to suggest that benthic barriers have little effect when placed on relatively coarse-textured sediments (Table 1)

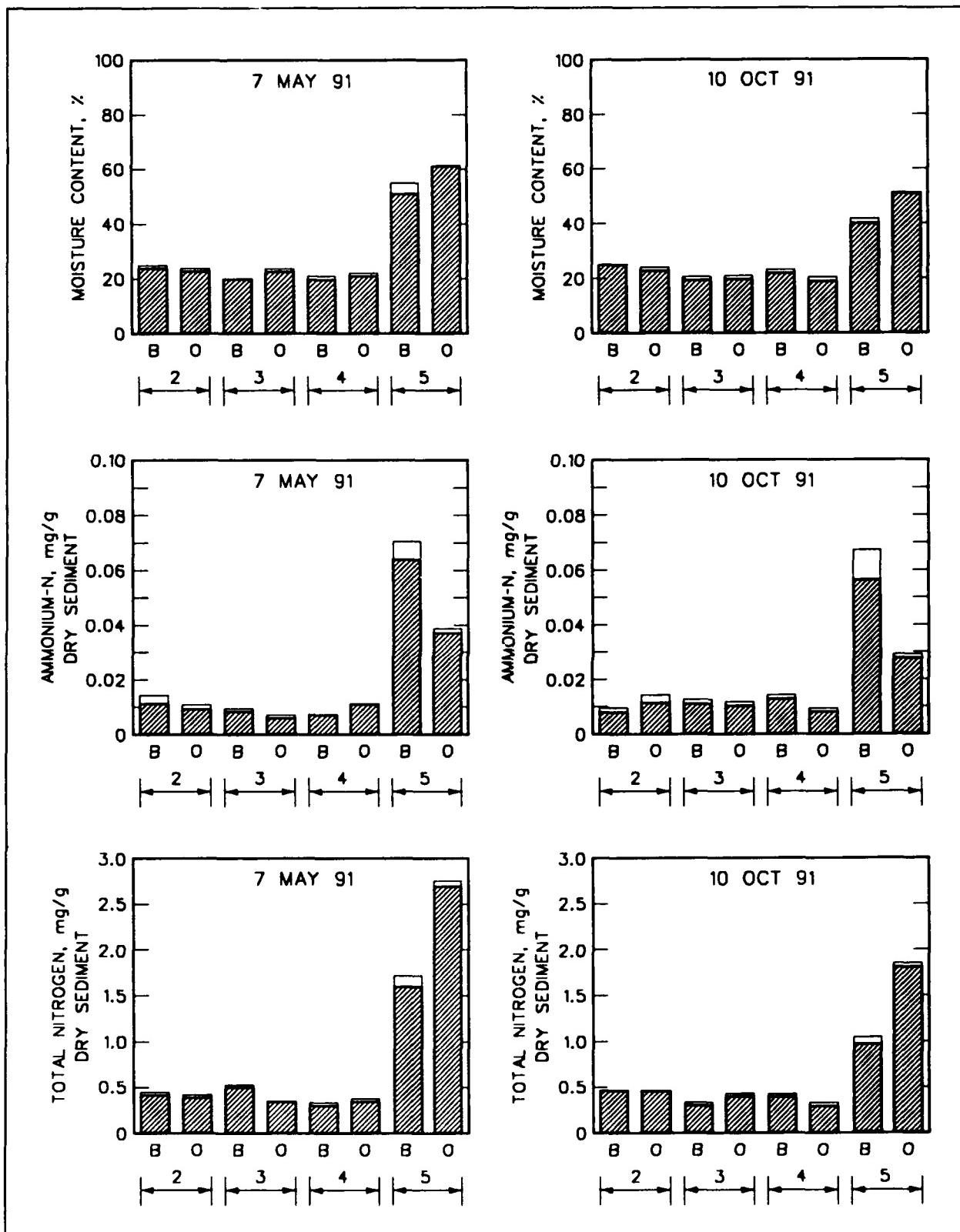


Figure 3. Means ($n = 3$) and standard errors of sediment composition for sediment collected beneath (B) and in the open (O) sediment adjacent to each barrier near the beginning and end of the plant growing season in Lake Guntersville

Table 1

Physical and Chemical Characteristics of Lake Guntersville Sediment Collected at Locations Beneath (Barrier) and Adjacent (Open) to Benthic Barrier Near Beginning and End of Plant Growing Season

Characteristic	Location	May 7, 1991		October 10, 1991	
		Sites 2-4 ¹	Site 5 ²	Sites 2-4 ¹	Site 5 ²
Total Sediment					
Texture, %					
Sand	Barrier	64.7 (3.5)	20.0 (1.3)	58.8 (3.6)	15.0 (3.4)
	Open	63.3 (3.2)	22.5 (1.5)	54.2 (3.9)	24.3 (1.7)
Silt	Barrier	21.0 (1.9)	55.0 (1.4)	22.0 (2.3)	62.1 (2.9)
	Open	22.8 (1.8)	55.7 (1.4)	28.4 (3.6)	49.1 (2.2)
Clay	Barrier	14.3 (2.5)	25.0 (1.4)	19.2 (1.4)	22.9 (0.6)
	Open	13.9 (1.7)	21.8 (1.6)	17.5 (1.3)	26.6 (0.8)
Density, g/m ³	Barrier	1.36 (0.02)	0.72 (0.10)	1.32 (0.02)	0.87 (0.04)
	Open	1.29 (0.04)	0.43 (0.02)	1.31 (0.06)	0.61 (0.01)
Moisture, %	Barrier	21.5 (0.5)	47.7 (4.1)	23.2 (0.6)	39.5 (2.2)
	Open	22.2 (1.0)	62.0 (0.7)	22.4 (0.8)	53.0 (0.3)
Organic matter, %	Barrier	2.7 (0.3)	6.1 (0.6)	2.5 (0.0)	4.8 (0.3)
	Open	2.5 (0.2)	9.2 (0.2)	2.6 (0.0)	6.9 (0.1)
Total Kjeldahl nitrogen mg/g ³	Barrier	0.42 (0.03)	1.60 (0.13)	0.37 (0.02)	1.03 (0.07)
	Open	0.37 (0.02)	2.69 (0.09)	0.39 (0.01)	1.85 (0.04)
Phosphorus, mg/g ³	Barrier	0.399 (0.037)	0.782 (0.111)	0.220 (0.022)	0.461 (0.017)
	Open	0.414 (0.037)	1.062 (0.027)	0.230 (0.011)	0.750 (0.042)
Extractable Nutrients					
Ammonium-N, mg/g ³	Barrier	0.01 (0.00)	0.06 (0.01)	0.01 (0.00)	0.06 (0.01)
	Open	0.01 (0.00)	0.04 (0.00)	0.01 (0.00)	0.03 (0.00)
Phosphate-P, mg/g ³	Barrier	0.010 (0.002)	0.072 (0.009)	0.013 (0.001)	0.071 (0.005)
	Open	0.012 (0.001)	0.100 (0.006)	0.017 (0.002)	0.130 (0.008)
Potassium, mg/g ³	Barrier	0.02 (0.00)	0.05 (0.00)	0.03 (0.01)	0.03 (0.00)
	Open	0.03 (0.00)	0.07 (0.00)	0.02 (0.01)	0.06 (0.00)
Interstitial Water					
Ammonium-N, mg/L	Barrier	1.49 (0.44)	6.87 (1.51)	1.44 (0.25)	6.80 (0.72)
	Open	1.21 (0.38)	1.94 (0.30)	0.96 (0.22)	1.51 (0.11)
Phosphate-P, mg/L	Barrier	0.067 (0.032)	0.380 (0.091)	0.009 (0.001)	0.148 (0.042)
	Open	0.072 (0.043)	0.600 (0.077)	0.007 (0.001)	0.293 (0.025)
Iron, mg/L	Barrier	5.5 (1.8)	25.9 (7.6)	4.1 (2.1)	19.0 (2.7)
	Open	3.0 (1.7)	9.3 (0.6)	3.3 (1.4)	5.9 (0.7)
Manganese, mg/L	Barrier	7.2 (1.7)	9.5 (2.2)	6.1 (1.2)	5.0 (0.8)
	Open	5.4 (0.5)	8.9 (0.4)	3.1 (0.6)	4.0 (0.5)

¹ Values are grand mean ($n = 9$) with associated standard errors.

² Values are grand mean ($n = 3$) with associated standard errors.

³ Based on sediment dry mass.

having low moisture and organic matter content, such as represented by sites 2-4. However, barriers placed on more finely textured sediments with higher levels of moisture and organic matter, as represented by site 5, do appear to affect sediment composition. Possible mechanisms by which benthic barriers affect sediment composition include sediment

consolidation, reduced chemical diffusion into the overlying water column, increased anoxia, and cessation of nutrient inputs to the sediment via sedimentation.

At Eau Galle Reservoir, no appreciable changes in physical and chemical composition of sediment collected from beneath the barrier

were evident over the macrophyte growth season, from June until September 1991 (Table 2). However, significant changes did occur in sediment collected adjacent to the barrier. Changes noted were in sediment texture, from a predominantly silt and clay composition (about 56 and 27 percent, respectively) to a sand and silt (about 56 and 33 percent, respectively) composition; in moisture content (from about 78 to about 54 percent); in density (from 0.16 to 0.59 g/ml); and in organic matter content (from 13.5 to 6.1 percent). These changes suggest a significant change in sediment type due to erosional and/or transport processes (James and Barko 1990).

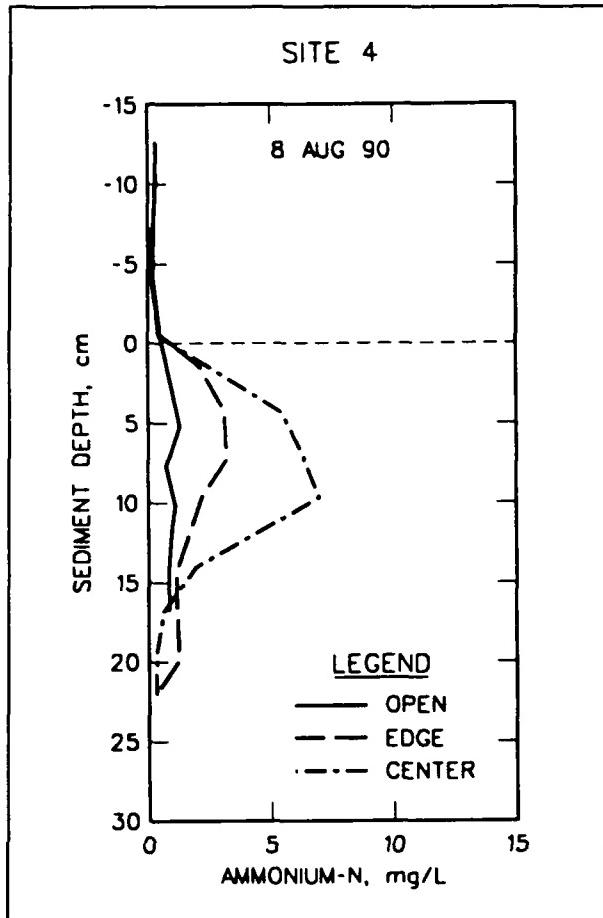
Interstitial water

Results from the pilot experiment at Lake Guntersville in 1990 showed NH₄-N concentrations in the interstitial water to increase near the center of each barrier (Figure 4). The gradient in NH₄-N observed from the edge to the center of the barrier may have been caused by greater water exchange or diffusional effects near the barrier's edge.

Throughout the study, profiles of NH₄-N, Fe, and Mn measured beneath the center of the barriers at Lake Guntersville exhibited consistently higher values as opposed to outside the barriers at all sites. Moreover, NH₄-N, Fe, and Mn concentrations beneath the barrier at site 5 were significantly higher than observed at sites 2-4 (Table 1). Seasonal influences of barrier placements were suggested by the greater differences in NH₄-N concentrations (Figure 5) that were seen, beneath the barrier and the open location, toward the end of the growth seasons (cf. September 1990, 1991, and June 1991) when temperatures, and probably microbial activities, as well, were higher.

Surficial water

Dissolved oxygen concentrations in the surficial water collected beneath barriers



*Figure 4. Profiles of interstitial NH₄-N concentrations collected *in situ* near the center, along the edge, and in the open sediment adjacent to the benthic barrier at site 4 in Lake Guntersville*

were consistently near zero. These levels tend to suggest that barrier permeability was insufficient to permit exchanges of dissolved oxygen from the overlying water column. Surficial water anoxia, observed at both Lake Guntersville and Eau Galle Reservoir, is considered a primary factor for the absence of viable benthic invertebrate populations.¹

Summary and Conclusions

Sediment conditions beneath benthic barrier placements at Lake Guntersville and Eau Galle Reservoir exhibited some physical and

¹ Personal Communication, July 1991, Barry Payne, US Army Engineer Waterways Experiment Station, Vicksburg, MS.

Table 2

Mean (n = 3) Physical and Chemical Characteristics (with Associated Standard Errors) of Eau Galle Reservoir Sediment Collected at Locations Beneath (Barrier) and Adjacent (Open) to Benthic Barrier Near Beginning and End of Plant Growing Season

Characteristic	Location	June 6, 1991		September 19, 1991	
Total Sediment					
Texture, % ¹					
Sand	Barrier	26.5		19.1	
	Open	17.3		55.6	
Silt	Barrier	51.4		61.2	
	Open	55.9		32.7	
Clay	Barrier	22.0		19.7	
	Open	26.7		11.7	
Density, g/ml ²	Barrier	0.20	(0.02)	0.27	(0.08)
	Open	0.16	(0.02)	0.59	(0.12)
Moisture, %	Barrier	73.6	(2.7)	74.9	(6.1)
	Open	78.2	(2.4)	53.9	(7.7)
Organic matter, %	Barrier	11.5	(0.8)	10.9	(1.6)
	Open	13.5	(0.7)	6.1	(1.1)
Total Kjeldahl nitrogen mg/g ³	Barrier	4.42	(0.20)	3.64	(0.29)
	Open	5.50	(0.23)	1.75	(0.33)
Phosphorus, mg/g ²	Barrier	1.362	(0.045)	1.081	(0.047)
	Open	1.738	(0.035)	0.518	(0.075)
Extractable Nutrients					
Ammonium-N, mg/g ²	Barrier	0.17	(0.00)	0.25	(0.03)
	Open	0.28	(0.03)	0.14	(0.03)
Phosphate-P, mg/g ²	Barrier	0.005	(0.002)	0.032	(0.011)
	Open	0.009	(0.004)	0.034	(0.007)
Potassium, mg/g ²	Barrier	0.20	(0.01)	0.20	(0.03)
	Open	0.26	(0.02)	0.13	(0.03)
Interstitial Water					
Ammonium-N, mg/L	Barrier	16.37	(0.99)	22.60	(2.21)
	Open	24.73	(1.47)	35.00	(2.33)
Phosphate-P, mg/L	Barrier	0.610	(0.061)	0.065	(0.036)
	Open	1.150	(0.511)	0.288	(0.179)
Iron, mg/L	Barrier	13.6	(1.9)	13.8	(1.3)
	Open	16.0	(3.1)	26.4	(6.0)
Manganese, mg/L	Barrier	0.2	(0.0)	3.2	(0.1)
	Open	0.3	(0.1)	4.2	(0.5)

¹ Based on a composite of three replicate samples.

² Based on sediment dry mass.

chemical modification. Greatest changes in both physical and chemical composition occurred on fine-textured sediments having relatively high moisture and organic matter contents (e.g. >30 and >3 percent, respectively). Reductions in both moisture and organic contents were most significant beneath the barriers placed over fine-textured sediments. Pore water-soluble nutrients, particularly NH₄-N, exhibited greater increases beneath barriers than adjacent to barriers.

Replenishment of nutrients to sediment beneath barriers via sedimentation inputs may have been severely restricted or eliminated because of barrier placement. Development of anoxic conditions in surficial water beneath barriers and subsequent eradication of viable benthic invertebrate populations occurred at all sites.

Although changes in aquatic habitat conditions do result from benthic barrier placements,

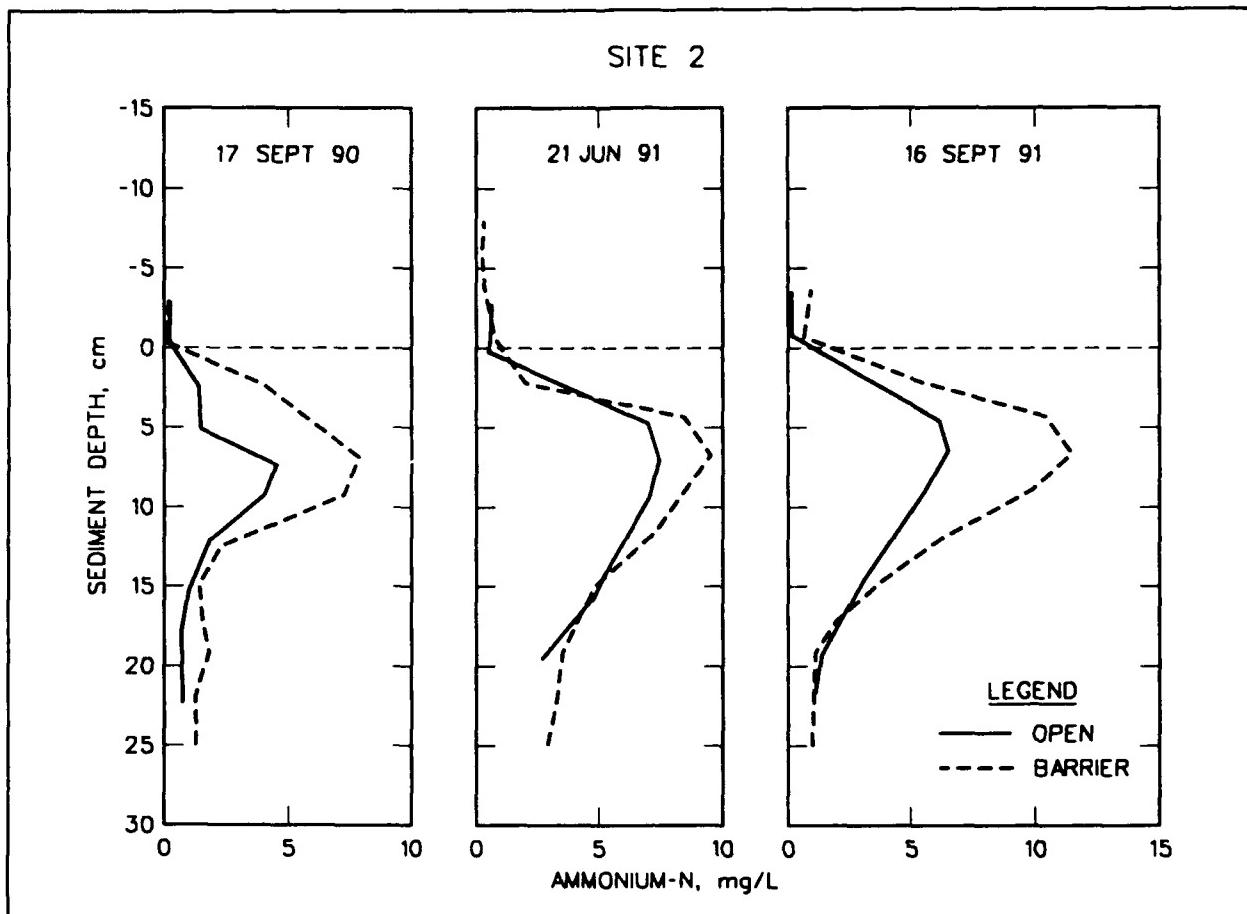


Figure 5. Profiles of interstitial NH₄-N concentrations collected *in situ* near the center and in the open sediment adjacent to the benthic barrier at site 2 in Lake Guntersville

these changes appear to be minor. Furthermore, it remains unclear whether these effects are detrimental to organisms other than macroinvertebrates. An examination of aquatic macrophyte growth on sediments collected from beneath benthic barriers is to be performed in an effort to ascertain any detrimental effects on their phenological or physiological condition.

Acknowledgments

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Production of Propagules of Native Aquatic Plant Species

by

Susan E. Monteleone¹ and R. Michael Smart¹

Introduction

In our studies of aquatic plant competition, we are evaluating the feasibility of establishing desirable native species in an attempt to slow the spread of weedy species. To conduct these experiments on a pond or lake scale, we require large quantities of propagules of selected native species. These experiments were conducted to determine efficient methods of production of native propagules. The two native species assessed were *Vallisneria americana* and *Potamogeton nodosus*.

Economical yet successful culture methods require minimal maintenance while providing maximum yield of propagules. Successful culture methods (a) provide sustained propagation by use of sexual and asexual reproduction, (b) ensure rapid biomass accumulation for sustained growth, and (c) take into account the ease of transplantation of species used in pond-scale studies.² Carl E. Korschgen and W. L. Green. 1988. American wildcelery (*Vallisneria americana*): Ecological considerations for restoration. Technical Report 19. Washington, DC: US Department of the Interior, Fish and Wildlife.

Sediment types and use of culture-medium nutrient supplementation are important factors to consider when designing experimental propagation methods. A sediment substrate should provide adequate nutrient resources for rapid biomass accumulation while facilitating efficient transplantation of specimens.

Pond sediment provides a naturally occurring sediment nutrient source, while sand, a nutri-

ent-deficient planting medium, requires continual addition of nutrients. However, root systems and winterbuds of plants anchored in sand are more easily removed intact and with minimal tissue damage than roots planted in fine-textured, clayey sediments. These considerations of planting medium are evaluated in a series of plant culture trials.

The objective of these experiments was to develop an efficient method for propagating native aquatic species such as *Vallisneria americana* and *Potamogeton nodosus* (pond-weed).

Methods

Raceway experiment

A factorial experiment was conducted with two water flow conditions (flowing or static) and three sediment types (sand that was nitrogen-amended with 1 g ammonium sulfate per 2-L container, pond sediment, and wetland sediment). Pond sediment was treated with metam-sodium, a soil fumigant, in an attempt to eliminate spores of *Chara vulgaris* and seeds of *Najas guadelupensis*.

Eight 2-L pots of each sediment type were planted with two *Vallisneria* specimens. Four replicates of each sediment type were randomly arranged in each raceway and subjected to either flowing or static conditions. After 50 days, plants (including both roots and shoots) were harvested, rinsed free of sediment and debris, oven-dried at 60 °C, and weighed to determine biomass accumulation.

¹ US Army Engineer Waterways Experiment Station, Lewisville Aquatic Ecosystem Research Facility, Lewisville, TX.

²

Pond experiment

Monocultures of two species, *Vallisneria* and pondweed, were planted in screened wooden frames (0.25 m^2 , approximately 9 cm deep) containing either pond sediment or sand. Sixteen specimens were planted in each frame. Four replicates of each species-sediment treatment were placed either on benthic barrier fabric or on bare pond bottom of Pond 29 at the Lewisville Aquatic Ecosystem Research Facility (LAERF). *Vallisneria* transplants were obtained from Toledo Bend Reservoir, Texas, while pondweed was planted from tubers collected at LAERF.

Before planting, pond sediments were treated with metam-sodium, a soil fumigant, to reduce spore and seed germination of invasive species. Plants were allowed to grow for 4 months in a natural pond setting before they were harvested, dried, and weighed to determine plant biomass. In addition to biomass, production of pondweed winter tubers was also determined.

Results and Discussion

Raceway experiment

Vallisneria produced maximum biomass on pond sediment and minimum biomass on wetland sediment (Figure 1). These differences were likely attributable to nutrient limitation of growth on the wetland sediment. Flowing-water conditions, with a constant influx of nutrient-rich lake water, resulted in increased growth of plants on the wetland sediment. However, flowing-water conditions had no effect on growth of *Vallisneria* on the nitrogen-amended sand. Likewise, flowing water did not improve the growth of *Vallisneria* on nutrient-rich pond sediment. In fact, flowing-water conditions favored the growth of endemic *Chara*, a nonrooted macrophytic algae that obtains its nutrients from the water column. The increased growth of *Chara* and *Najas* on the pond sediment held under flowing-water conditions may have caused a concomitant reduction in growth of *Vallisneria* under these conditions.

Nitrogen-amended sand yielded comparable *Vallisneria* biomass to that produced on pond sediment and did not suffer massive invasions by *Chara* and *Najas*. In addition, sand was easy to handle and is obtainable in large quantities. Plant roots were also easily removed from the sand substrate with minimal damage. As a planting medium, nitrogen-amended sand may prove to be a good choice for culturing of *Vallisneria*.

Pond experiment

Both *Vallisneria* and pondweed grown on pond sediment accumulated greater above- and below-ground biomass than those grown on sand (Figure 2). Both species grown on pond sediment placed on the barrier had the greatest total biomass accumulation. Only pondweed did well on pond sediments that were placed on bare pond bottom. On the pond bottom, *Vallisneria*, a low-growth form species, was overgrown by endemic *Najas* and *Chara*, while pondweed, a canopy-forming species, was not. Competition for light and nutrients by *Najas* and *Chara* may have inhibited the growth of *Vallisneria* on the pond bottom.

On the barrier, competition was not a factor. Growth of pondweed was reduced on sand, particularly on the barrier, presumably due to nutrient limitation. In addition to reducing competition from endemic species, the barrier also restricted nutrient exchange between the pond bottom and the planting frames.

Like biomass accumulation, pondweed tuber production was greater on pond sediment than on sand (Figure 2). Tubers were more numerous on sand placed on bare pond bottom (22) than sand placed on the barrier (2), which indicated that pondweed tuber production may have been limited on the barrier due to nutrient restrictions.

This set of experiments, in the raceway and in the pond, led to the conclusion that sand may be the best rooting medium, based on the fact that sand restricted invasion by endemic species while providing comparable plant growth to plants grown on nutrient-rich pond

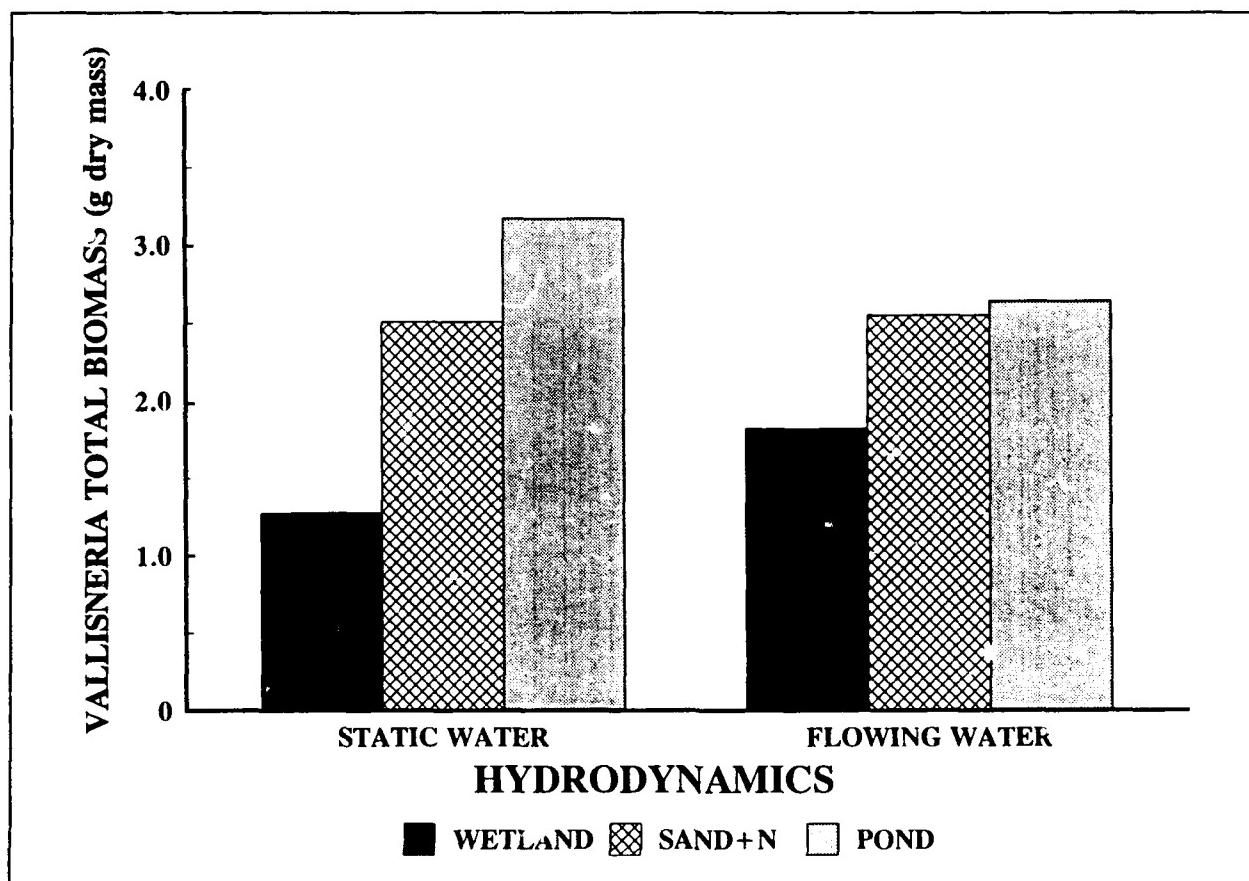


Figure 1. Total biomass (grams dry mass) of *Vallisneria americana* planted in flowing water and static water raceways on three different sediment types

sediment. Sand is nutrient poor, but a consistent source of nutrients may maximize plant growth during a long-term culturing experiment.

Long-Term Culturing Trial

The long-term culturing of *Vallisneria* is still in the experimental stages. Researchers have had some measure of success using both sexual and asexual means of propagation.^{1,2} We considered a rapid means of propagation to be the best course of action; thus, vegetative (asexual) propagation, the production of daughter plants via stolon shoots, was the culture method employed.

Based on the results of the previous experiments on culturing native species on nitrogen-amended sand, pond sediment, and wetland sediment, we were of the opinion that the ideal system for culturing native species would employ a coarse substrate to facilitate removal of the plant with minimal tissue damage and also to minimize competition from endemic species. However, coarse substrates are poor providers of nutrients and would require continuous additions of nutrients. We reasoned that nutrients provided in the rooting medium, rather than in the water column, would provide maximum growth of rooted macrophytes and minimal growth of undesirable algae. For these reasons we chose to

¹ C. E. Korschgen and W. L. Green. 1988. Technical Report 19. Washington, DC: US Fish and Wildlife Service.

² Personal Communication, S. A. Kollar, Harford Community College, Harford, MD.

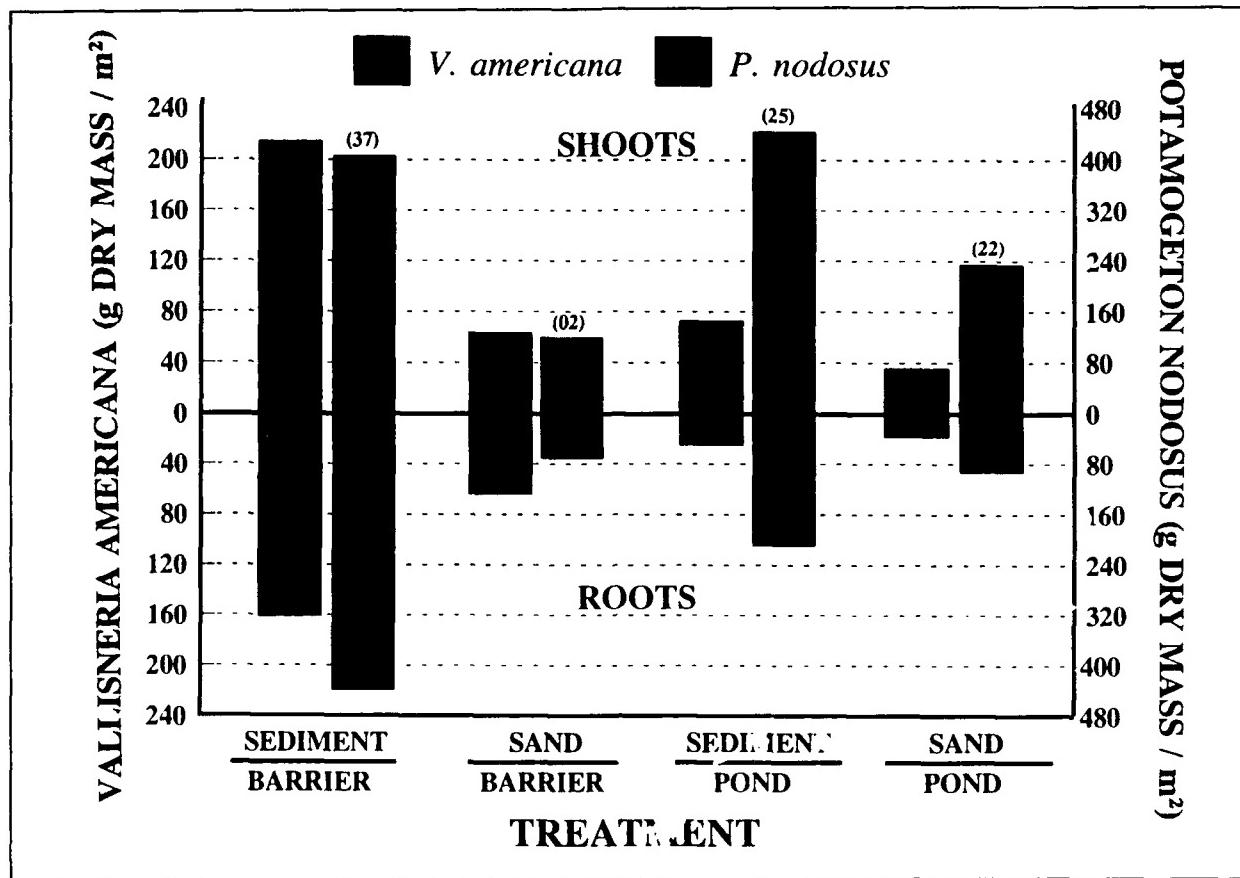


Figure 2. *Vallisneria americana* and *Potamogeton nodosus* shoot and root biomass (grams dry mass/m²) for each sediment type that was placed on benthic barrier fabric or bare pond bottom. Solid bars represent pond sediment planting medium and hatched bars represent sand planting medium. Average tubers per treatment are presented as values in parentheses.

establish *Vallisneria* in a bed of coarse sand with a subsurface flowing-water system.

The raceway was plumbed with perforated polyvinyl chloride (PVC) pipe overlain with

a bed of coarse sand (Figure 3). Fresh lake water is passed through the root system of the plants, reducing the potential for algal growth in the water column since most of the nutrients are removed as lake water passes

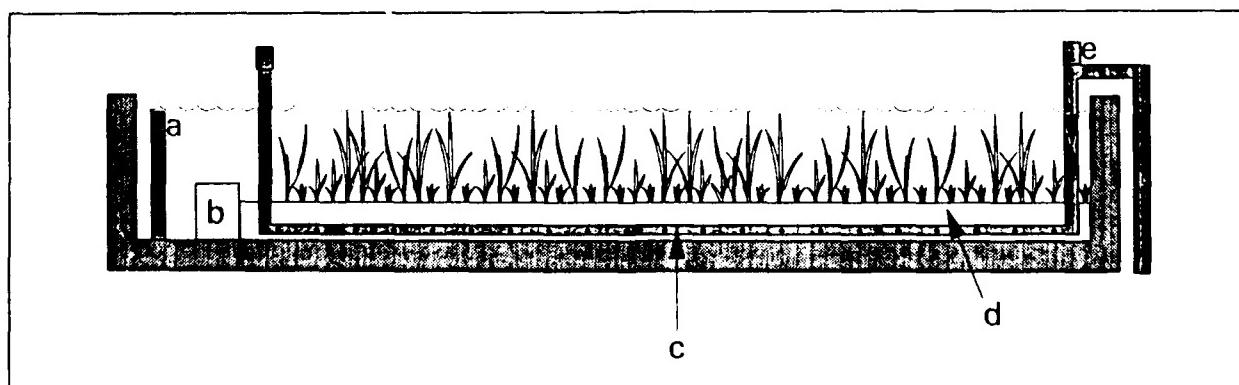


Figure 3. Experimental design for *Vallisneria* cultures in raceways (a—standpipe, b—cinderblock barrier, c—perforated PVC pipe, d—coarse sand, e—fill valve)

the roots. This root-flow system can also be supplemented with nutrient additions as needed.

Ongoing Research

Vallisneria plants have been growing in this system for several months, and we are continuing to observe and evaluate this method of culturing. We are experimenting with potting native species in peat containers to improve the efficiency of transplanting cultured species in pond-scale experiments. Potted *Vallisneria*

plugs could easily be removed from our flowing-water raceway culture bed and systematically planted in experimental ponds. If successful, this culture method may provide a more efficient system of propagation and transplantation of native aquatic species.

Acknowledgments

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Biological Control Technology

Biological Control Overview

by

Alfred F. Cofrancesco, Jr.¹

The fiscal year (FY) 1991 direct allotted biological research was apportioned among nine work units, described briefly below.

- *Biological Control of Hydrilla Using Insects.* The fly *Hydrellia pakistanae* has been established in Florida and is dispersing. This insect has also been released in Alabama, Georgia, and Louisiana, and adults have been collected away from the release sites. *Hydrellia balciunasi* was released in Texas, and adults were also collected away from the release site. *Bagous* n. sp. was released in Florida, and adults have been recaptured from the site.
- *Temperate Biocontrol Insects for Eurasian Watermilfoil and Hydrilla.* A number of potential biocontrol agents have been found in China that impact hydrilla. Some of the most promising are two *Hyrellia* flies, one of which was a population of *H. pakistanae* from the colder region around Beijing. The *H. pakistanae* population from China was brought into quarantine and is being screened, with the hope that it can be released quickly, because of the testing that has already been done on this species. The other fly is a new species, which Dr. Deonier will be describing. Some weevils have also been found on Eurasian watermilfoil, and they are being identified by Dr. O'Brien. These weevils are at the US Quarantine Facility in Florida for identification and testing.
- *Biological Control of Eurasian Watermilfoil Using Pathogens.* An Environmental Use Permit has been received from the US Environmental Protection Agency for use of the fungal pathogen *Mycoleptodiscus terrestris* (MT) as a microherbicide. The EcoScience formulation of MT was tested in ponds at Lewisville, TX, in FY 91.
- *Biological Control of Hydrilla Using Plant Pathogens.* A patent has been submitted for the fungal pathogen isolated from hydrilla in Texas. Greenhouse studies continue to test the efficacy and specificity of the pathogen.
- *Biological Control of Waterlettuce with Insects.* The weevil *Neohydronomous affinis* is well established in Florida and has caused a dramatic decline in the waterlettuce population at particular sites. This weevil has also been released in Louisiana and Texas. A second biocontrol agent, the moth *Namangana pectinicornis*, has been released in Florida at selected sites.
- *Biological Management of Aquatic Plants with Allelopathic and Competitive Species.* In greenhouse studies, three native plants exhibited allelopathy to hydrilla. Studies showed that *Ceratophyllum*, *Vallisneria*, and *Potamogeton* added to the soil caused a reduction in hydrilla biomass or height after 4 weeks.

¹ US Army Engineer Waterways Experiment Station, Vicksburg, MS.

- *Herbivorous Aquatic Insects on Eurasian Watermilfoil.* The surveys for insects on Eurasian watermilfoil have been completed in New England. Both Coleoptera and Lepidoptera adults and larvae were collected at various sites. Also, there were additional reductions in watermilfoil populations where large weevil populations were observed.
- *Hydrilla Biocontrol Insects in TVA.* Tennessee Valley Authority personnel are rearing *H. pakistanae* at Muscle Shoals, AL. Over 80,000 larvae were released at Lake Guntersville, Alabama, in 1991. Adult flies were collected away from the release cages, indicating that the population was developing and spreading. A significant reduction in the hydrilla mat occurred in the fish enclosure area where the flies were released.
- *Eurasian Watermilfoil Biocontrol Pathogens in TVA.* Surveys were conducted in Lake Guntersville, and a number of pathogens were collected. MT was isolated from the decline of Eurasian watermilfoil that was observed in the Kentucky Lake systems. The decline extended 45 miles along the system.

Release and Field Colonization of New Biological Control Agents of *Hydrilla verticillata*

by
Ted D. Center¹

Hydrilla, *Hydrilla verticillata* (L.f.) Royle (Hydrocharitaceae) is a submersed, caulescent vascular hydrophyte that is widely distributed in the Old World. Adventive infestations of this plant constitute the most severe aquatic plant problem in the southern United States (Schmitz 1990, Schmitz et al. 1991). Between 1982 and 1989 it infested 40,000 to 60,000 acres in Florida alone (Schardt and Schmitz 1989). During the same period it rapidly expanded its range into northern regions and now occurs as far west as California (Sonder 1979) and as far north as Delaware (Haller 1982).

Control of hydrilla is achieved primarily through the use of herbicides or herbivorous fish (Sutton and Vandiver 1986), but both methods are prohibitively expensive for widespread use. Development of alternative approaches has therefore been afforded high priority by Federal and State agencies. One possible alternative involves the introduction of host-specific plant-feeding insects obtained from within the native range of hydrilla. Accordingly, searches for these potential biological control agents began during the 1970s in India and Pakistan (Sankaran and Rao 1972; Baloch and Sana-Ullah 1974; Baloch, Sana-Ullah, and Ghani 1980) and resumed during the 1980s in other areas of the world (Center and Dray 1990; Center, Cofrancesco, and Balciunas 1990). The resultant surveys revealed the presence of numerous biological control candidates, including an undescribed *Hydrellia* species later named *H. pakistanae* Deonier (1978).

Hydrellia pakistanae is a small, ephydrid fly whose larvae mine hydrilla leaves. Baloch and Sana-Ullah (1974) and Baloch, Sana-

Ullah, and Ghani (1980) described the life history and biology of this species. Eggs are laid on floating leaves, and the larvae either enter the leaves directly or descend the water column before entering the plant. Females apparently oviposit indiscriminately, but the larvae are quite fastidious (Buckingham, Okrah, and Thomas 1989). Each larva destroys about 12 leaves during the course of its development and then pupates within a puparium attached in a leaf axil (Baloch and Sana-Ullah 1974; Buckingham, Okrah, and Thomas 1989). Total generation time is 18 to 30 days (Baloch and Sana-Ullah 1974).

Laboratory studies done in Pakistan (Baloch, Sana-Ullah, and Ghani 1980) and later in a US quarantine facility (Buckingham, Okrah, and Thomas 1989) confirmed that *H. pakistanae* was host-specific and suitable for release in the United States. Accordingly, Dr. Gary Buckingham petitioned the Animal and Plant Health Inspection Service (USDA-APHIS-PPQ) Technical Advisory Group (TAG) on Biological Control of Weeds in June 1987 for permission to release this species. Permission was obtained and the first field release of *H. pakistanae* was made on 29 October 1987 (Buckingham 1988a,b; Center 1989).

During surveys conducted in Australia, a second *Hydrellia* species was discovered attacking hydrilla (Balciunas 1987; Balciunas and Center 1988; Balciunas, Center, and Dray 1989). Although the species was unknown at the time, it was later named *H. balciunasi* (Bock 1990). Further study in Australia and at Dr. Buckingham's laboratory confirmed that it, too, was completely host-specific and safe for release in the United

¹ USDA-ARS, Aquatic Plant Control Research Unit, Fort Lauderdale, FL.

States. After appropriate documentation, it was approved by APHIS on 9 May 1989.

Australia also provided a highly damaging stem-boring weevil from hydrilla (Balciunas 1987; Balciunas and Center 1988; Balciunas, Center, and Dray 1989). This unnamed species (initially misidentified as *Bagous australasiae*) is presently being referred to as *Bagous* n. sp. Z (the "Z" was appended so as to distinguish this species from other new species in the same genus).

The adults feed externally on the leaves and stems of hydrilla, causing the stems to fragment. Single eggs are laid in the stems, usually in or near a leaf node. Larvae burrow within the stems, causing extensive tissue destruction and further fragmentation. When the larvae mature, they apparently seal off a stem fragment and float to shore in it. They pupate on the shoreline, usually within a distinct strand line. After emerging as adults, they fly back to the hydrilla beds (see Balciunas and Purcell, in press).

In this report we document the release of these three species and the establishment of self-perpetuating field populations of *H. pakistanae* and their initial dispersal to other sites. This project represents the first attempt to control a submersed aquatic plant species using the "classical" or "introduction" approach to biological control.

Methods and Materials

Hydrellia pakistanae— the Asian hydrilla fly

Buckingham, Okrah, and Thomas (1989) documented the collection of the original *H. pakistanae* stock from India and its receipt and disposition at the Gainesville, FL, quarantine facility during the period May to August 1985. These researchers also developed an effective rearing procedure in which eggs were placed in 1-gal jars containing hydrilla. As adults emerged, they were transferred to an egging chamber (sleeve cage) that contained hydrilla sprigs in shallow pans of

water. The adults oviposited on the exposed portions of the hydrilla sprigs, and the egg-laden sprigs were then placed into the jars to perpetuate the rearing process.

After permission was granted for release of *H. pakistanae*, a portion of the quarantine colony was used for the first release, a portion was retained in quarantine, and a portion was transferred to our field laboratory in Fort Lauderdale, FL. Essentially the same rearing procedures were followed in Fort Lauderdale. Stock was later provided to the Waterways Experiment Station (WES) and to the Florida Department of Natural Resources (FDNR) Bureau of Aquatic Plant Control in Tallahassee, FL. The colonies at WES continued to be reared in the manner described above, with minor modifications. The stock cultures sent to FDNR were placed into a large, hydrilla-filled pool housed in a glasshouse.

The first release of *H. pakistanae* took place at Lake Leonore (also called Lake Patrick) near Frostproof in Polk County, Florida, on 29 October 1987 using insects acquired from the quarantine colony. Only one release was made at this site, and it consisted of a mixture of eggs, larvae, and adults. Because of the distance of the site from both Gainesville and Fort Lauderdale, it was revisited infrequently. The first follow-up examination was conducted on 29 January 1988, and the site was not visited again until the following October, after learning that the hydrilla population had collapsed the previous August.

The Fort Lauderdale colony eventually increased to a size capable of yielding sufficient insects for field release. The first insects from this culture were released in southern Florida during February and March 1988. These first liberations consisted exclusively of eggs that were attached to hydrilla sprigs. We obtained the eggs by removing adults from the culture jars, placing them in egging chambers with hydrilla sprigs for 2 to 3 days, counting the eggs, and then transferring the egg-laden sprigs to the field site. The sprigs were usually placed among leafy stems in the upper layer of the existing hydrilla infestation

which, in most cases, had grown to the surface of the water.

Early attempts at establishing field colonies of *H. pakistanae* were unsuccessful. Although we were unsure of the reasons, we suspected that the numbers released were inadequate for the simultaneous emergencies of reproductive adults that were needed to produce a breeding population. We also feared that predation by flying insects, particularly damselflies, might limit the reproductive life span of the few adults that managed to survive. Additionally, coots and moorhens sometimes selectively ate the sprigs of hydrilla that we had placed at the sites, thereby reducing or eliminating the initial stock. We therefore modified our procedures.

First, we began to release larvae rather than eggs. We felt that 7- to 10-day-old larvae would have a better chance of survival because, in the lab at least, mortality rates tended to be highest for first instars. Eggs were obtained in the same manner as before, but the sprigs were held in the laboratory prior to release for 10 to 14 days. Second, we increased the numbers of insects released per site. We accomplished this at first by improving our rearing methods and intensifying our rearing efforts, so as to produce more insects. Later, after field colonies were established, we supplemented the laboratory-reared adults in the egg boxes with field-collected adults, thus greatly increasing the size of the releases. We reduced the number of sites at which we made releases and we chose small, local sites that we could visit frequently. This enabled us to release insects repeatedly and at frequent intervals until we could conclude that they had either established or that establishment was not possible. By releasing at smaller sites, we increased the number of insects per unit of surface area and thereby improved our chances for early recovery of adults if establishment occurred.

We also instituted the use of shallow, floating cages to contain the plant material harboring the larvae. These cages were constructed of two square frames made of 1-in.-

square, channeled aluminum tubing. The frames were attached to one another with a piano hinge along one edge. The top side of the upper frame was covered with fine-mesh screen, but the bottom frame was unscreened. The hollow aluminum tubes were filled with polystyrene to provide flotation. The cages were placed at each site with the screened side up, and were held in place with poles anchored into the hydrosoil that passed through U-bolts attached to the outside of the cage. Access into the cage was provided by lifting the hinged upper frame. The insects were released into the open lower frame.

The cages served several functions. Although they did not confine the insects completely, they did help retain them within a small area, thereby increasing the likelihood that emerging adults would encounter one another and form mating pairs. The cages also clearly demarcated the point of release, which facilitated our ability to recover flies from successive generations and to verify establishment. Also, because the cages were shallow (2 cm freeboard), maneuverability by flying predators was very limited, thus decreasing predation and improving adult survival. The cages also served to protect the plant material harboring the insects from herbivores such as moorhens, coots, and ducks.

Each time a new release was made, the site was checked for the presence of adult flies both inside and outside the cage. Occasionally, if insects seemed established in the cage but could not be found outside of it, the cage was moved and further releases were made at a new locus. Releases were discontinued only after we became convinced that the additional numbers were insignificant relative to the number of flies that were already present. We considered *H. pakistanae* to be established only if we consistently recovered adults for five generations, or approximately 4 months.

In early 1991, after *H. pakistanae* populations were established at several sites, we initiated surveys to determine if populations had dispersed to other areas. This involved

visiting most release sites, as well as other hydrilla localities, and collecting a sample of *Hydrellia* spp. from the site. Flies were collected by pushing a floating sheet of white polystyrene along the water surface and catching the flies that landed on it. A hand vacuum that had been adapted for this purpose was used to catch the flies. We generally collected about 100 adults, which we immediately preserved in isopropanol. We examined the sample at the Fort Lauderdale laboratory and sorted the specimens by species. Voucher specimens were prepared, and the identity of the first specimens was verified by Dr. R. L. Deonier of Miami University, the taxonomic expert on *Hydrellia*.

Material from the FDNR colony was released at a few sites. In these instances the fly-infested plant material was removed from the pool and transferred directly to a field site. No attempt was made to quantify the numbers or stages released.

On 25 August 1990 Dr. Buckingham received a consignment of field-collected *H. pakistanae* from Kumbaleagodu, Bangalore, Karnataka State, India. He received another consignment from Rawalpindi, Pakistan, on 28 August 1990. These were placed in culture jars and reared as before. However, Dr. Buckingham held these colonies for only 60 to 70 days (two to three generations), while identifications were confirmed, the purity of the material was checked, and parasites were eliminated.

On 30 October 1990, portions of these colonies were transferred from the quarantine facility to the field laboratory. We released the insects from the Pakistani colony (an estimated 1,000 larvae and pupae plus 334 adults) on the same day in northern Florida at the Little River just east of the Wacissa River in Jefferson County near Tallahassee. The remainder were released in southern Florida at two small catchment ponds located within the Florida Turnpike rights-of-way in northern Broward County (designated the Lakeview sites). The two ponds were on opposite sides of a road from one another. The Indian

strain was released only at the northern pond, and the Pakistani strain was released only at the southern pond. The Indian strain was first released on 5 November 1990 (ca. 1,000 larvae and pupae).

We received additional material of both strains from the Gainesville colonies on 6 and 14 November 1990. All of these were released at the two ponds on 14 November 1990. The total quantity released in the north pond equaled ca. 1,400 larvae and pupae plus the uncounted contents of four culture jars (probably <100 larvae and pupae per jar). The quantity released in the south pond was less, totaling only 200 larvae and pupae plus the contents of one culture jar.

Hydrellia balciunasi— the Australian hydrilla fly

The first consignments of *H. balciunasi* were from North Pine Dam, Petrie, Queensland, Australia, and were received in US quarantine on 3 February 1988. Buckingham (1989) completed host testing in quarantine and submitted documentation to the TAG on 2 November 1988. After an appropriate review, the TAG granted permission to release this insect on 5 May 1989. Colonies were transferred from Gainesville to Fort Lauderdale shortly thereafter. Large colonies were developed during the ensuing few months using the same procedures then in use for *H. pakistanae*.

Hydrellia balciunasi was first released at a small pond (the Orangebrook Country Club site) in Broward County, Florida, on 1 August 1989, using the cages as described above. By April 1990 we had released about 12,000 eggs and larvae by making 27 separate releases into small enclosures at weekly intervals (Table 1). A single adult *H. balciunasi* was recovered about 25 m from the enclosure on 7 December 1989, so we assumed that it had established at the site. Although we continued to release at the site, later collections of hundreds of adults provided only *H. pakistanae* which, to our knowledge, had never been released there. Further checking

Table 1
Releases of Hydrilla Leaf-Mining Fly (*Hydrellia balciunasi*) in Florida

Site	County	Release Dates	Number Releases	Number Released				Infested Sprigs ²	Status ³
				Eggs	Larvae	Pupae	Adults ¹		
Orangethorpe Golf Club	Broward	9/89-4/90	27	1,127	>10,086	1,376	U	Yes	P, ⁴ HP
Wilson Cypress Strand	Broward	3/91-5/91	3	+ ⁵	9,717 ⁵	0	0	No	I,HP
Big Gant Lake	Sumter	8/91	1	0	0	0	0	Yes	I
Lake Panasofkee	Sumter	4/91-12/91	5	+ ⁵	5,1375	+ ⁵	0	Yes	I
Naples Manor	Lee	11/91	1	0	1,066	0	0	No	I,HP

¹ A designation of U indicates members of the stage in question were known to be in the material released, but actual counts were unavailable.

² Affirmative responses indicate plants were released from cultures that harbored unknown quantities of *H. balciunasi*.

³ A designation of P indicates a weak positive indication, HP indicates that *H. pakistanae* had overwhelmed the site, and I indicates it was too early or we were unable to evaluate the site.

⁴ By 9 February 1990, our *H. balciunasi* cultures had become contaminated with *H. pakistanae*. Thus, subsequent releases at this site contained unknown numbers of *H. balciunasi* and *H. pakistanae*.

⁵ Various stages released but not separately counted. Totals listed under larvae.

revealed that our *H. balciunasi* colonies were contaminated with and overwhelmed by *H. pakistanae*, and this had been true for some time. We later learned that the colonies at the Gainesville quarantine laboratory (which provided our stock) were also contaminated. We therefore purged our colonies by releasing the remaining material at the site.

We later received "clean" stock from Gainesville, and established new pure colonies of this species. However, these colonies never flourished and produced only marginal quantities of flies. A total of about 20 adults, 194 larvae, and 83 eggs were shipped to WES personnel to assist in establishing a second colony there. The further removal from our colony of about 500 eggs and larvae by WES personnel in Texas further depleted our colonies, which failed to recover and ceased production within a few weeks. Fresh stock was then obtained from Australia, and new colonies were established both in Fort Lauderdale and in Vicksburg. Although these new colonies have only recently begun to produce sufficient material for release, we continued to release this species in Florida using insects supplied from the WES colonies.

Bagous n. sp. Z— *the hydrilla stem weevil*

Studies on the biology and host specificity of *Bagous n. sp. Z* were initiated in Australia

by Dr. Balciunas in 1986. Data obtained were sufficient to request permission to bring weevils into quarantine. Dr. Buckingham did so on 4 September 1986. Approval was obtained later that year, and the first consignment from Australia entered the quarantine facility on 15 April 1987.

Extensive testing both in quarantine and in the field in Australia provided grounds to request permission to allow this insect to be released from quarantine. Dr. Buckingham transmitted a request for this permission on 5 October 1989, and permission was granted on 2 February 1991. Colonies were first transferred to Fort Lauderdale during April 1987, and the first insects were released on 8 March 1991.

The first release consisted of small numbers of adults and larvae that were placed at Lake Osborne in Palm Beach County, Florida. The site appeared to be satisfactory because of the presence of hydrilla in extensive strand lines along the shore of the lake. Data from Australia indicated that these strand lines might be necessary pupational sites for this species of weevil (Balciunas 1987; Buckingham 1988a, 1988b, 1989, 1990; Balciunas, Center, and Dray 1989). Infested plant material containing the larvae was incorporated into the stranded hydrilla. The adults were released both on the shoreline and on the surface of submersed hydrilla beds.

Additional releases have now been made at the Orangebrook Country Club pond in Broward County and Lake Panasofkee in Sumter County. The insect releases at Orangebrook were derived from insects shipped directly from Australia. The adults were the F1 progeny of weevils collected at North Pine Dam near Petrie, Queensland, between 3 and 18 April 1991. The larvae were F2 progeny of the original adults. These were released immediately upon receipt from Gainesville in order to avoid any undue laboratory rearing. The weevils were first released (15-16 July 1991) in a 1- by 1-m cage similar to those used for *Hydrellia* releases. The cage was situated so as to extend from the hydrilla bed in the water to the soil at the shoreline. On 17 July the square cage was replaced with a longer rectangular cage (2.3 m by 0.9 m by 5.1 cm), which more effectively straddled the shoreline.

Our original laboratory colony provided the weevils released at Lake Panasofkee. The first release was made on Spring Brook, a creek that flows into the south end of Lake Panasofkee. The site was about midway between the mouth of the lake and the I-75 bridge over the creek. Later releases were made below the I-75 bridge under the north-bound lane in a open area that had been cleared for construction of the bridge. The hydrilla was growing between and beneath the bridges up to the treeline. The hydrilla beds near the treeline were exposed during periods of low water. The bridge provided shade, which excluded emergent vegetation so that nothing was present on the exposed muck. The weevils were placed on hydrilla in semidry and wet, mucky areas under the cover of the bridge.

Results and Discussion

Hydrellia pakistanae— the Asian hydrilla fly

Early releases. When we first began to release *H. pakistanae* we were unable to recover it during later site examinations. However, we could not confidently conclude that field

populations had not established. We thought that perhaps small, localized populations had established at levels too low to detect. Later experience allayed this fear. We found that when populations do establish, flies quickly become abundant and specimens are easily collected. This knowledge reinforced our confidence in our ability to judge whether the various release attempts were successful. These data are summarized in Table 2.

Hydrellia pakistanae was first released at Lake Leonore in Polk County, Florida, during October 1987. During fall 1987 the extent of the hydrilla infestation on Lake Leonore was estimated at 300 acres and was at problem levels (i.e., growing to the lake surface). When we examined the site in January 1988, we failed to recover *H. pakistanae*.

During September 1988, a FDNR biologist (Mr. Terry Sullivan) visited Lake Leonore to estimate the extent of the hydrilla infestation as part of an annual plant survey. He discovered almost a total lack of hydrilla at the water surface. Although he estimated that about 200 acres remained, it was deemed to be at nonproblem levels, mostly confined to the bottom. He later learned from Polk County aquatic plant control specialists that the hydrilla infestation had collapsed during the previous July or August. After further checking, he confirmed that no control measures had been implemented at the lake, and nothing comparable had happened at other lakes in the area. With the knowledge that hydrilla beds that reached the water surface had been present there year after year, he suspected that this sudden demise was the result of the release of *H. pakistanae* the year before (T. Sullivan, personal communication).

Mr. Sullivan contacted the Fort Lauderdale laboratory on 29 September 1988, and within a few days (3 October 1988) we met with him at the site. We collected hydrilla from the lake bottom that was completely brown and deteriorated but with green sprigs growing from a few nodes. Decomposition was too extensive to ascertain the cause of the decline. We collected 45 *Hydrellia* spp.

Table 2

Releases of Hydrilla Leaf Mining Fly (*Hydrellia pakistanae*) in Florida and Southern Georgia

Site	County	Release Dates	No. Re-releases	Number Released ¹				Infested Sprigs ²	Status ³
				Eggs	Larvae	Pupae	Adults		
Lake Leonore ⁴	Polk	10/87	1	6,000	2,600	0	600	No	N
Rodman Reservoir at Spike Club	Marion	11/87	1	1,000	0	0	0	No	N
Everglades Holiday Park ⁴	Broward	2/88-6/88	2	6,177	0	0	0	No	P
Lake Hicpochee ⁴	Glades	3/88-5/89	3	5,093	550	0	0	No	N
Lake Hicpochee ⁵	Glades	3/90-9/90	25	0	31,319	578	0	No	P
Lake Osborne	Palm Beach	5/88	1	3,870	0	0	0	No	I
Fisheating Bay, L. Okeechobee	Glades	8/88-3/89	2	387	3,078	0	0	Yes	N
Big Bear Beach, L. Okeechobee	Glades	11/88-11/90	6	9,749	215	0	1,034	No	I
Eagle Bay Island, L. Okeechobee	Okeechobee	11/90-12/90	14	120,359	650	U	3,000	Yes	E
North New River canal	Broward	1/89	1	0	0	0	0	Yes	I
Lake Tohopekaliga	Osceola	3/89-12/90	3	3,227	143	0	3,300	No	I
Sears Lake ⁴	Polk	3/89	2	3,531	0	0	0	No	N
US27/SR78 canal	Glades	4/89	1	410	119	0	0	No	I
Palm Beach Airport pond ⁴	Palm Beach	11/89-1/90	7	13,223	3,140	157	0	No	E
Lake Seminole Fishing lodge ⁴	Decatur, GA	6/90-7/91	15	138,376	23,921	747	3,544	Yes	I
Flint River ⁴		6/90-9/90	4	19,308	5,875	688	3,244	No	N
Kidney Slough		10/90-11/90	3	2,040	6,905	59	0	No	N
Spring Creek		5/91-7/91	5	111,069	0	0	300	No	I
		6/91-7/91	3	5,959	11,141	0	0	No	I
Hacienda Village pond ⁴	Broward	1/90-2/90	9	18,582	10,249	0	0	No	E
Wacissa River System	Jefferson	10/90-11/90	34	10,430	2,661	1,841	3,834		
Horsehead Run ⁴		10/90	51	10,430	1,561	153	3,500	No	I
Boat ramp spring		11/90	13	0	600	600	0	Yes	I
River run 1 mi S of springs ⁴		10/90	1	0	0	588	0	Yes	P
Little River		10/90	9	0	500	500	334	Yes	I
Lakeview (north pond) ⁴	Broward	11/90	2	0	650	650	0	Yes	I
Lakeview (south pond) ⁴	Broward	11/90	1	0	100	100	0	Yes	E
Lake Robin Hood	Lake	12/90	1	0	0	0	1,200	No	I
Orangebook Golf Club pond ⁶	Broward	2/90-7/90	10	U	U	U	U	Yes	E
Total			111	340,414	79,395	4,073	16,512		

¹ A designation of U indicates that actual counts were unavailable.² Infested plants, usually the contents of culture jars, harboring unknown quantities of *H. pakistanae*.³ A designation of E indicates definite establishment, P designates possible establishment, N designates no indication of establishment, and I designates indefinite results.⁴ Site at which hydrilla declines were observed after release of *H. pakistanae*.⁵ An *H. pakistanae* population persisted briefly at this site prior to elimination of the hydrilla infestation by flushing.⁶ Originally an *H. balciunasi* release site, but *H. balciunasi* cultures were adulterated by *H. pakistanae*. Thus, some or all releases made at this site contained both species. Only *H. pakistanae* established at the site.

adult flies from the surfaces of yellow water-lily pads and other floating plant material, but most proved to be *H. bilobifera*, a closely related native species. Two specimens appeared to be *H. pakistanae*, but upon closer examination proved to be a third species (*H. discursa*). While there, we also collected four samples of hydrilla. These were held and checked periodically for several days. No *H. pakistanae* emerged. We concluded that the collapse of the hydrilla population was not caused by *H. pakistanae*.

The site was later checked on 2 March 1989 and 28 March 1989. The hydrilla beds had recovered and were at or near the surface in the release areas. No *H. pakistanae* were recovered. We last examined the site on 27 July 1990 and collected extensively with Dr. R. L. Deonier. A total of 91 *Hydrellia* spp. specimens were obtained, which Dr. Deonier determined to be four species, but none were *H. pakistanae*. We have concluded that *H. pakistanae* was never established at this lake.

A similar pattern occurred at Lake Hicpochee. Flies were released there in March 1988, but no ensuing effort was made to determine if a population established or persisted. The mat on which the release was made all but disappeared by fall. Nearby mats persisted, however, indicating that whatever had impacted the mat at the release site was only locally active. Again, we had no indication that *H. pakistanae* populations had previously established at the site nor that they were responsible for the decline.

Flies were also released in Everglades Holiday Park, where a population temporarily established and had begun to disperse. A small hydrilla bed located across a canal from an airboat concession was chosen as the first release site. Flies were released on 26 February 1988, and a single adult *H. pakistanae* (from six *Hydrellia* spp. specimens) was recovered on 26 April. A second release was made on 28 June in an area very near to the first. Six specimens of adult *Hydrellia* flies were collected at the original release site on

1 September, but none proved to be *H. pakistanae*.

By 22 September the original hydrilla bed was no longer present at the point of release, but extensive beds persisted elsewhere in the area. At that time, hydrilla samples were collected from three areas within the vicinity of the release sites. One *H. pakistanae* was reared (emerged 4 October 1988) from a sample collected from an old hydrilla bed directly across the canal from the original release point. About 100 m west of the site, two *H. pakistanae* adults (from nine *Hydrellia* spp. specimens) were field-collected, and one adult was reared from a plant sample (emerged 4 October 1988).

We eventually collected a total of 25 *Hydrellia* spp. adults in the area, but only the two *H. pakistanae*. Again, although the only hydrilla bed to disappear was at the initial release site, we have little evidence to connect the two events. Later, most of the remaining hydrilla died out due to drought conditions in the area.

Sudden collapses of hydrilla populations proved to be commonplace. The results of our efforts were confounded by such declines at Sears Lake, at Lake Seminole, and at the two Lakeview ponds, as well as at Lake Leonore, Lake Hicpochee, and Everglades Holiday Park. Insufficient data exist to associate this phenomenon with the release of *H. pakistanae*, but the coincidence is becoming difficult to explain.

Numerous releases made during the 2-year period between October 1987 and late 1989 brought the cumulative totals released to about 44,000 eggs, 4,600 larvae, and 600 adults. Despite initial recoveries of small numbers of the releases made before 1990 produced field populations of *H. pakistanae*. One obvious potential problem was the extensive length of time that the flies had been bred in captivity before being released. We suspected that we had produced a laboratory strain that was poorly adapted to field conditions. We

therefore initiated the necessary procedures to acquire fresh stock from overseas. However, this proved to be a lengthy process, and consignments were not shipped until over a year later. In the meantime, we continued to work with the existing stock.

We identified several other possible problems. We assumed that we were releasing too few flies at too many sites. Also, the sites, many of which comprised hundreds of acres, were much too large. The presence of vast expanses of hydrilla at these sites precluded easy verification of establishment (i.e. the "needle in the haystack" scenario). Also, we noted that eggs were easily dislodged from the hydrilla sprigs and that mortality was highest for first instars. We therefore held them longer prior to release to allow time for the eggs to hatch and the larvae to grow. After we began to repeatedly release large numbers of later instars into caged enclosures at small sites, we noticed the populations persisting.

Later releases. The first bona fide establishment of *H. pakistanae* occurred at three small ponds in southern Florida. One was a drainage pond for a newly constructed interstate highway (the Hacienda Village pond), the second was a borrow pit located at the West Palm Beach International Airport, and the third was a golf course pond (the Orangebrook pond) located in Hollywood.

We released mainly immature stages into cages on the surfaces of the "topped-out" hydrilla beds at each site. We released as many insects as we could procure as often as possible, frequently at weekly intervals. Nine releases were made at the Hacienda Village site during January and February 1990, which consisted of nearly 29,000 eggs and larvae. Seven releases were made at the airport site between November 1989 and January 1990, totaling over 16,000 eggs, larvae, and puparia.

Populations established at both sites within 2 months. Insects released at the golf course pond were initially presumed to represent a different species (*H. balciunasi* Bock). How-

ever, after initial recoveries of a few *H. balciunasi*, subsequent collections provided only *H. pakistanae*. This was traced back to contamination by *H. pakistanae* of some *H. balciunasi* cultures while in quarantine; thus, instead of successfully colonizing a second *Hydrellia* species, we established an additional population of *H. pakistanae*.

Lake Hicpochee. Encouraged by the results at the small sites, we then attempted to establish the flies at a larger site. We chose Lake Hicpochee for this purpose. Lake Hicpochee is located at the southwest end of Lake Okeechobee. The lake is bisected by the Caloosahatchee River. We selected the section on the north side of the river as a release area and began a release program during March 1990. This continued until September 1990, during which time we made 25 releases totaling over 30,000 insects (Table 2). (We also made three releases consisting of about 5,000 insects during 1989, but these failed to establish because of the collapse of the hydrilla population.)

Unfortunately, when the site was revisited on 7 September 1990, the hydrilla was gone. Almost all of the aquatic vegetation appeared to have been scoured out by a massive movement of water through the site. A few small patches of hydrilla were found, however, and a few adult *H. pakistanae* were collected from them. Thus, we felt that a population of *H. pakistanae* probably was established in the area. We included the site in our later survey and collected a sample of flies on 27 June 1991. Even though only a small amount of hydrilla was present, 13 *H. pakistanae* were collected (eight females, five males) from a total of 135 *Hydrellia* spp.

Lake Okeechobee. Lake Okeechobee contains the largest hydrilla infestation in Florida, with over 8,000 acres infested (Chardt and Schmitz 1989). Thus, we were of the opinion that, if we could establish *H. pakistanae* there, its continued survival in Florida would be assured. We therefore expended considerable effort to achieve this objective.

The first attempts at Fisheating Bay and Big Bear Beach made prior to 1990 were not successful. We then focused our efforts in the area around Eagle Bay Island. The first release made at this site was on 21 November 1990 and consisted of about 3,000 adult flies. We collected these from the Orangebrook site during the preceding 2 days and held them in a sleeve cage provisioned with hydrilla. The flies were transported in the sleeve cage to the site and released directly onto the surface of a bed of hydrilla in an open area surrounded by bullrush. The contents of 21 culture jars were also placed at the site.

On 6 December 1990, a cage was placed on the hydrilla bed about 10 m north of the point of the first release, and 6,648 eggs and larvae from the WES culture were placed in it. An additional 16,923 eggs and larvae from the WES culture were placed in the cage on 21 December 1990.

By 22 January 1991 we noticed that adult flies were present in the cage, so we moved it about 100 m to the east. About 18,400 eggs from the WES culture were released at the new location. Also, the contents of 42 culture jars were placed at the location of the earliest release. Four additional lots totaling over 80,000 eggs and larvae from the WES culture were placed in the cage between 28 February and 22 March 1991. Adult flies were collected in the area on 7 March (43 flies) and 15 March 1991 (14 flies), but none proved to be *H. pakistanae*.

Then, on 22 March 1991, we collected 85 adult flies, 24 of which were *H. pakistanae* (14 females, 10 males). Most of these were found near the earliest point of release where we had placed adult flies and the contents of culture jars onto the hydrilla in the open.

On 10 May 1991 another sample of 85 flies included 15 *H. pakistanae* (5 female, 10 males). We checked the site on 11 July 1991, at which time we collected a sample of 14 flies. Seven of these were *H. pakistanae* (five males, two females), but populations

were sparse because of the deterioration of the hydrilla beds.

We last checked the site on 19 August 1991 and collected a sample of 17 *Hydrellia* spp. Only three of these were *H. pakistanae*, and the deterioration of the hydrilla beds had progressed further. Curiously, this deterioration was evident only in the area where the flies had been released. Plenty of healthy hydrilla beds persisted in adjacent areas. We believe that *H. pakistanae* populations are established in the vicinity of Eagle Bay Island and have probably dispersed into other portions of Lake Okeechobee.

Wacissa River System. The Wacissa River in Jefferson County, Florida, originates at a series of springs located about 24 km southwest of Tallahassee. The water in the river is clear and cool, averaging about 21 °C (Rosenau et al. 1977).

We released *H. pakistanae* in four areas of this system: Horsehead Run between Horsehead Spring and Log Spring; the spring adjacent to the boat ramp at the park; the Wacissa River about 1.5 km below Horsehead Run; and Little River about 0.75 km northeast of its confluence with the Wacissa River. The *H. pakistanae* placed at each location were from various sources.

On 31 October 1990, we released about 3,500 field-collected adults from the Orangebrook site in Horsehead Run. We then moved downstream and emptied the contents of 71 culture jars into a cage at the second site. This included at least 588 puparia and uncounted larvae from the Fort Lauderdale cultures. We then proceeded up the Little River a short distance, where we released 334 adults plus about 1,000 larvae and puparia of the Pakistani strain. On 7 November 1990, we released about 400 larvae and puparia of the Pakistani strain at the second site and 800 larvae and puparia of the Indian strain at the Little River site. On 15 November 1990, we released 400 larvae and puparia of the Indian strain at the first site and 200 larvae and puparia at the second site.

On 3 January 1991, we collected a sample of flies from the site. We failed to keep these separated, but most were collected at the second site in the main river run. From the 30 flies collected, we obtained one *H. pakistanae*.

On 22 February 1991, we moved hydrilla that had been infested with *H. pakistanae* from the FDNR greenhouse to a small spring adjacent to the boat ramp. We also collected samples of flies from the three original release sites. The 38 flies collected in Horsehead Run included three female and one male *H. pakistanae*. The five flies collected at the second site included one female and two male *H. pakistanae*. The 57 flies collected at the Little River site included a male and a female *H. pakistanae*.

Despite some indication of establishment, we continued to release in the system. We released about 23,000 eggs at the second site on 29 March 1991 and another 8,200 on 15 April 1991, and about 15,000 eggs from the WES colony in the boat ramp area on 8 April 1991, all from the WES colony. We again collected samples of flies on 3 May 1991 and found one male and one female *H. pakistanae* at the boat ramp (from 71 *Hydrellia* spp. collected); none at the Little River site (from 36 specimens collected); and none at the second site (from 69 collected). No hydrilla existed in Horsehead Run.

We again collected samples of flies on 18 October 1991. The samples were as follows: 68 from near the boat landing; 4 in Horsehead Run; 108 from the river; and 61 at Little River. None of these 240 flies were *H. pakistanae*. Thus, we tentatively conclude that the population has disappeared from the area, although we cannot rule out the possibility that they might persist elsewhere in the system.

Lake Seminole. The release site at Lake Seminole was located in Georgia on the north side of the lake across the channel from Wingate's Fishing Lodge in a protected cove behind a small grass island. Three cages were placed at the site. We made an initial

release of 289 puparia and about 1,500 larvae in a single cage on 26 June 1990. On 23 August we installed two additional cages and released 84 puparia and 2,715 larvae from Fort Lauderdale and WES cultures into the original cage; 1,727 field-collected adults from the Hacienda Village site into the second cage; and 5,168 eggs from the field-collected adults in the third cage. The cages were oriented in a line and spaced about 50 m apart.

At the time of the first release (26 June 1990), the hydrilla was quite healthy. However, water levels were low as a result of the prevailing drought conditions in the area. This resulted in an abnormal exposure of the hydrilla beds to solarization. Later when the second release was made (23 August 1990), water levels had increased but the hydrilla beds had deteriorated significantly. Nonetheless, an additional 1,238 eggs from the WES culture were released into the original cage on 19 September 1990.

On 26 September 1990 the hydrilla bed below the second cage was nearly gone, so we moved the cage nearer the shore onto healthier plants. We placed material from field-collected flies (from Hacienda Village) into all three cages, including 1,660 larvae and 315 puparia in the first cage, 1,727 adults in the second cage, and 12,902 eggs in the third cage.

Deterioration of the hydrilla worsened over the ensuing period, and on 16 October 1990 we decided to abandon the site. The third cage was then moved to a healthier hydrilla bed located nearer the main channel in the Flint River. However, only three releases were made in this cage near the end of the growing season. The first release, made on 16 October 1990, consisted of 2,020 eggs, 1,533 larvae, and 59 puparia from both the Fort Lauderdale and WES colonies. The second and third, made on 8 and 16 November 1990, consisted of 3,657 larvae and 1,715 larvae, respectively, from the WES colonies.

The site was checked again the following spring (2 May 1991). The hydrilla beds in

the original release areas had recovered, but those in the alternate site were in poor condition, apparently having been scoured out by flooding. We collected extensively in both areas and failed to recover *H. pakistanae*.

We then abandoned both sites and moved one cage to an entirely different location in Kidney Slough on Spring Creek. At the same time, we released 32,164 eggs from the WES culture into the cage. Later releases from the same source included 36,525 eggs on 15 May, 26,740 eggs on 23 May, and 15,640 eggs on 4 June. On 20 June we installed a new cage in the Spring Creek area and released 5,959 eggs from the WES colony into it.

On 10 July we released 300 field-collected adults and 1,530 larvae at the Kidney Slough site. The adults were collected at the West Palm Beach site on 5 October and held in an egging chamber. The larvae were progeny that hatched from their eggs. We released another lot of 9,611 larvae from the WES colony at the Spring Creek site on 18 July 1991. Five additional consignments containing 3,574, 5,137, 3,584, 1,379, and 2,930 larvae, all from the WES colony, were placed into the cage at Spring Creek at weekly intervals between 4 September and 9 October 1991.

Examination of the hydrilla within the cage on 18 October 1991 uncovered 23 empty puparial cases, suggesting that some of the larvae had survived and ultimately emerged as adults. However, an alligator had been regularly climbing on top of the cage to sun itself. This repeatedly submerged the cage, probably drowning the entrapped adult flies. Therefore, a new, taller cage was constructed and placed in the same area. One release of 2,776 larvae from the WES colony was made into this cage on 23 October 1991.

We collected samples of flies from the original release area on 2 May 1991, prior to moving the cage to the Spring Creek area. None of 66 specimens taken were *H. pakistanae*. We then collected in the vicinity of the Flint River releases, and *H. pakistanae* was not found among 55 flies collected. We also collected 141 flies from two sites near

Spring Creek and, not unexpectedly, found no *H. pakistanae*. On 10 and 18 July 1991 we collected 58 and 59 flies, respectively, at Kidney Slough; 55 and 62 flies at a nearby cove; and 43 and 44 flies in the original release area. No *H. pakistanae* were found.

On 18 October 1991 we collected 11 adults at Kidney Slough and, although no *H. pakistanae* were included, two *H. pakistanae* were included among seven flies collected in the Spring Creek area. Both of these were males; one was collected in the cage, but the other was collected from the hydrilla beds outside the cage. Hence, despite an intensive effort, we have been forced to conclude that *H. pakistanae* is not yet established at Lake Seminole. However, these recoveries from the Spring Creek area offer encouraging signs that a population is in the beginning stages of establishment.

Lakeview ponds. When the Pakistani and Indian strains of *H. pakistanae* were released at the two Lakeview ponds, hydrilla occupied about 75 percent of the surface of the north site and about 40 percent of the surface of the south site. On 29 January 1991 the hydrilla was nearly gone at both sites, but substantially more persisted at the south site. In both cases hydrilla extended to the surface only around the edges of the ponds. About 10 percent of the surface was occupied at the north pond as compared to about 20 percent at the south pond. Only two adult flies were collected on the hydrilla at the north site, and neither one was *H. pakistanae*. Fifteen flies were collected on the hydrilla at the south site, twelve of which (five females, seven males) were *H. pakistanae*. Hence, it appears that the Pakistani strain readily colonized the south site despite the small numbers released (ca. 200 larvae and puparia and the contents of one culture jar). It is possible that the Indian strain also established but was lost when the hydrilla population collapsed. This suggests that perhaps fresh material collected overseas and released directly in the field with a minimum amount of laboratory rearing may indeed establish more readily.

We cannot, however, legitimately compare the two strains on the basis of this experience. We later collected *H. pakistanae* at both ponds but, by then, we could not be sure that the Pakistani strain from the south pond had not colonized the north pond or that populations from other sources had not moved into the area.

Dispersal of *H. pakistanae*. We successfully established *H. pakistanae* populations in Florida at the West Palm Beach Airport pond, the Hacienda Village pond, the Orangebrook Country Club pond, Lake Hicpochee, Lake Okeechobee, the two Lakeview ponds, and possibly at the St. Mark's River. Populations also may be established in Lake Seminole, Georgia; Guntersville Reservoir, Alabama; and in southern Louisiana.

While collecting plant material for our laboratory colonies, a population of *H. pakistanae* was also found at a small land-locked lake (L Lake) located in Davie about 3 km south of the Hacienda Village pond. We noticed that flies were emerging from cultures sooner than expected, and we suspected that they were being introduced with the plant material. We examined freshly collected hydrilla on 12 February 1990 and found puparia. These were isolated in petri dishes, and adult *H. pakistanae* emerged on 21 February 1990. Additionally, we placed hydrilla in 10 culture jars while at the site. These jars were not inoculated with eggs. We then isolated these in a screenhouse well from any other cultures or source of flies. Adult *H. pakistanae* emerged in these jars also on 21 February 1990. Hence, although flies were not intentionally released at this lake, a population was clearly present.

Later, on 13 March 1991, while attempting to find an isolated site for the release of *H. balciunasi*, we collected a sample of flies from a site located about 100 km west of Fort Lauderdale. A total of five flies were collected from the surface of the hydrilla beds, and all proved to be *H. pakistanae*. The near-

est known *H. pakistanae* field populations were the Hacienda Village pond 90 km to the east and Lake Hicpochee 70 km to the north.

This site was again checked on 27 March 1991, at which time we collected 30 adult flies, but no *H. pakistanae* were found. We also collected at another site 9 miles farther west. There we collected 78 adult *Hydrellia* spp., which included five *H. pakistanae* (three females, two males). On 16 May and 14 June 1991 we collected 117 and 583 *Hydrellia* spp., respectively, at the first site and 33 (16 females, 17 males) and 275 (161 females, 114 males) were *H. pakistanae*.

The *H. pakistanae* populations that we discovered west of Fort Lauderdale existed in a canal that parallels the main east-west highway (Alligator Alley, State Route 84). The Hacienda Village pond lies adjacent to the same road, and a nearly continuous canal system connects the two locations. Although hydrilla populations are not continuous and many barriers occur along the way, it is conceivable that the Hacienda Village pond was the source of this population. We continued to survey farther west and recovered *H. pakistanae* at locations as far west as Naples, about 150 km west of the Hacienda Village pond (Figure 1).

Our next discovery of an adventive *H. pakistanae* population was at Lake Osborne in Palm Beach County. A total of 97 *Hydrellia* spp. were collected on 14 May 1991. This sample included six *H. pakistanae* (four females, two males). This lake is about 9 km south of the West Palm Beach airport pond. The airport pond is land-locked, but it lies near a continuous system of canals and lakes that includes Lake Osborne. Although Lake Osborne was an original release site, no flies had been released there since May 1988 and, despite several attempts, we were unable to substantiate that these releases were successful. We therefore feel that this site was colonized by flies that originated at the airport pond.

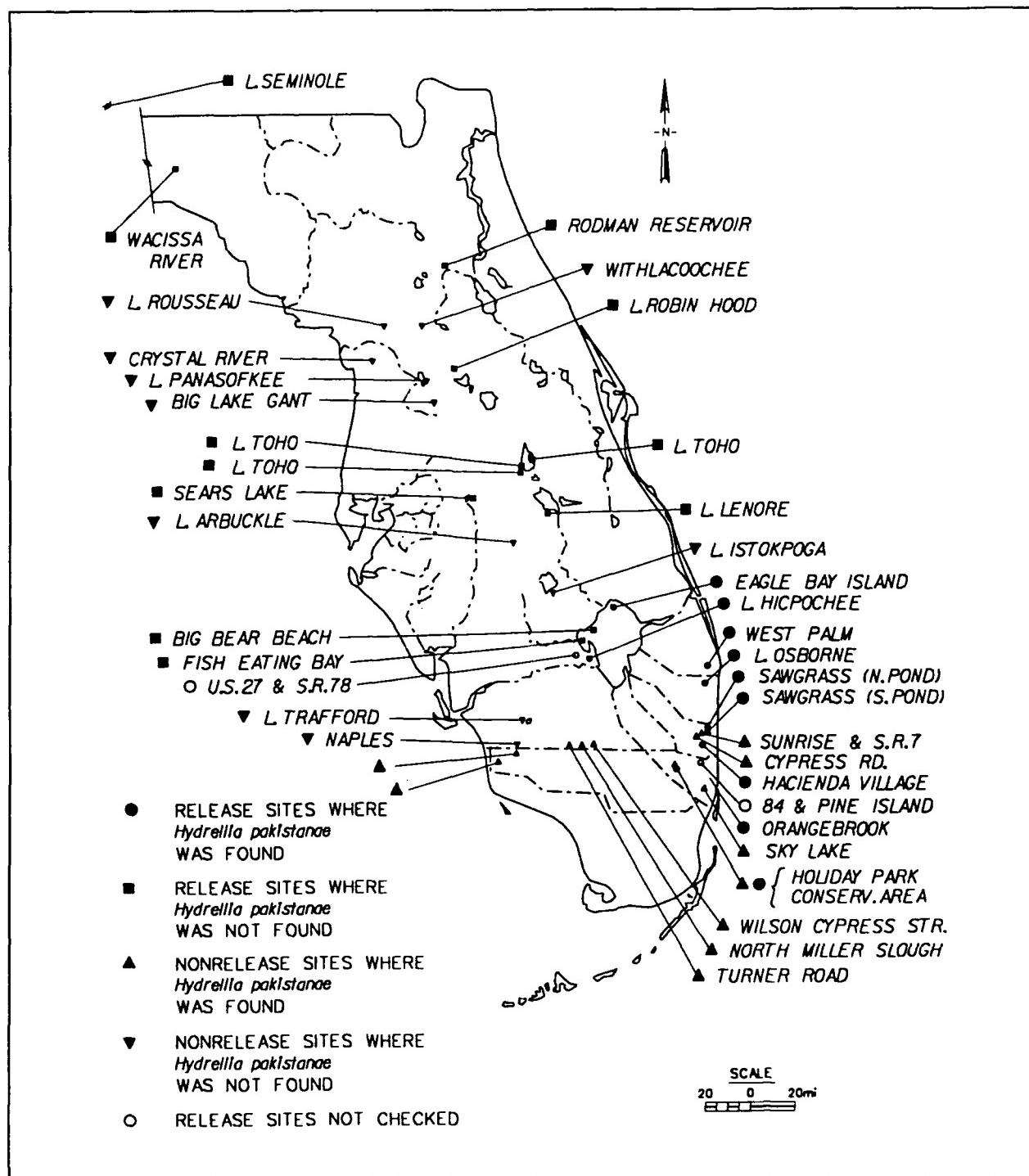


Figure 1. Map of Florida showing release sites and the present, known distribution of *Hydrellia pakistanae*

Hydrellia balciunasi— the Australian hydrilla fly

Because we have heavily committed to maintaining colonies of the hydrilla stem weevil and the waterlettuce moth, and because a

thriving, viable *H. balciunasi* colony has now been established at WES, we have not committed to the production of large numbers of *H. balciunasi*. Instead, we are presently maintaining only a small colony of this insect for release in Florida. Preliminary reports on

attempts to establish *H. balciunasi* in Texas have been encouraging (M. Grodowitz, WES, personal communication), so early establishment in Florida may not be critical.

Rather than undertaking further labor-intensive rearing efforts, we hope soon to be able to collect flies from field sites in Texas. Also, results of release attempts in Florida have been confounded by the increasingly ubiquitous presence of *H. pakistanae*. For example, after finding that the Orangebrook site contained mainly *H. pakistanae*, we began to search for alternate sites in which to release *H. balciunasi*. We found a good site on 13 March 1991 about 100 km west of Fort Lauderdale on Alligator Alley (SR 84) at a point nearly intermediate between Lake Hicpochee and Everglades Holiday Park. The site consisted of a hydrilla bed that existed in a cypress slough (Wilson Cypress Strand) adjacent to the road. This site was very isolated, so we felt that it would be safe from adulteration by *H. pakistanae*.

Initially we released 12,550 eggs and larvae. These were placed into the open hydrilla beds rather than into a cage. Before releasing these insects, however, we collected five *Hydrellia*-like flies from the surface of the water above the hydrilla bed. These were later identified, as noted above, as *H. pakistanae*. Despite the occurrence of low numbers of *H. pakistanae*, we continued to release *H. balciunasi* at the site (Table 1) until 16 May 1991. By then, *H. pakistanae* had become abundant and, even though *H. balciunasi* might have persisted there, we felt that further efforts to establish this site for use as a field colony were futile. A total of 19,434 eggs and larvae were placed at the site.

Later sampling failed to recover any *H. balciunasi*, although *H. pakistanae* remained abundant. Flies were collected at the site five times during the period 13 March to 27 November 1991. A total of 989 specimens were obtained of which 641 (65 percent) were *H. pakistanae*. The bulk of the remainder were *H. bilobifera*. *Hydrellia balciunasi* were recovered at the site only twice in ex-

tremely low numbers (seven specimens). Six of the seven specimens were collected on 27 March 1991, only 2 weeks after the release of 12,550 eggs and larvae. One was collected on 14 June 1991, about a month after the previous release.

Recognizing that it would be difficult to find a site that was free of *H. pakistanae* anywhere south of Lake Okeechobee, we began to look for a site in central Florida. We ultimately settled on two lakes near Tampa—Big Lake Gant and Lake Panasofkee. We began releasing *H. balciunasi* at those locations in April 1991 (Table 1). We also made one release (in November 1991) at a site in Naples, but it too was infested by *H. pakistanae*. We have not yet recovered *H. balciunasi* from any of these sites and cannot yet conclude that it is established in Florida.

Bagous n. sp. Z— the hydrilla stem weevil

Because we have only just begun working with this insect, it would be premature to draw conclusions regarding its establishment in Florida. However, some results have been encouraging. Only three releases were made at Lake Osborne, with only 1,082 adults and 23 larvae. The last release was made on 28 June 1991. Two weevils were recovered 70 days later, on 6 September 1991. These weevils were found by Dr. Balciunas during a recent visit to Fort Lauderdale. The weevils that were recovered showed no signs of wear on their vestiture and appeared to be recently emerged. Hence, they seemed to represent at least F1 progeny of the released insects. Unfortunately, later efforts failed to duplicate this success.

Weevils were released at the Orangebrook site between 15 July and 24 July using F1 and F2 progeny of insects collected in Australia during the previous April. The site was checked repeatedly during the first few weeks after the weevils were released. Adults were found almost every time, either in the cage or near the cage until 2 August. However, later efforts were again fruitless.

No weevils have ever been recovered from Lake Panasofkee. Because of the distance to the site, we have been able to visit the site only about once a month. Because we know that the weevils have persisted in the field for substantial durations, we are optimistic that populations are established. We hope that they may have merely moved away from the release areas to more suitable locations. If so, they should be more easily found as populations increase in size. In the meantime, we intend to devote most of our efforts during 1992 toward establishing this insect and are now working to increase the number and size of our colonies.

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Temperate Biocontrol Insects for Eurasian Watermilfoil and Hydrilla

by

Gary R. Buckingham¹

This travelogue portion of the program is usually presented by Dr. Joe Balciunas, the aquatic plant foreign explorer. Unfortunately, he is currently at the USDA/ARS Biocontrol of Weeds Laboratory, in Townsville, Australia, and was unable to return for this meeting. For the past three summers, Dr. Balciunas has traveled in China searching for insects that attack hydrilla and Eurasian watermilfoil, or milfoil (*Myriophyllum spicatum* L.). Results of his surveys can be found in the previous two proceedings. At his request, I traveled in China last year and in China and Korea this year visiting some of his sites later in the year and surveying in new areas.

Dr. Balciunas visited China this year from 1 to 29 July. He was based again, as was I, at the Sino-American Biological Control Laboratory (SABCL), Chinese Academy of Agricultural Science, in Beijing. He and SABCL researchers met with cooperators in Shenyang, Liaoning Province (3-7 July), and in Hohhot, Inner Mongolia (14-19 July), to discuss sampling procedures and to visit their sampling sites. The previously discovered milfoil weevil *Phytobius* sp. 1 was common, but no new insect species were found. A new area surveyed this year (7-20 July) was near the city of Harbin, Hei-long-jiang (Heilungkiang) Province, north of Beijing near Russia. The milfoil weevil was common. Larvae of another weevil were recovered from hydrilla carried to Beijing. They appeared to be *Bagous* larvae, but adults will need to be collected or reared to confirm this.

Korea

I began my trip to China in Korea where the USDA/ARS has a biological control laboratory. I was there for one week (11-19 August) to determine if hydrilla and Eurasian watermilfoil were common and if they had insects feeding on them. My host in Korea was Dr. Robert Pemberton, who is currently studying natural enemies of pest insects, for example, the gypsy and apple ermine moths. Dr. Pemberton surveyed in Africa in 1976 for hydrilla insects. He thus has both experience and interest in aquatic weeds. We visited 15 to 20 sites within a half day's drive of Seoul. About half of the sites had hydrilla and/or Eurasian watermilfoil, which was less abundant than hydrilla. However, even at the heavily infested hydrilla sites, the plants were in rather discrete clumps and the populations were restricted to small areas, unlike the situation in Florida where entire lakes are infested by dense mats of hydrilla.

Leaf-mining flies, *Hydrellia* sp., were found at all hydrilla sites, but they were not abundant. The puparia, or resting stage, was heavily parasitized by small wasps. Tip midges (larvae of small, mosquito-like, non-biting flies) were common on hydrilla at several sites; 60 to 80 percent of the tips were infested. Milfoil tips were also attacked by a midge, but that was much less common. No weevils were found on the plants during my brief visit. Striking damage was observed on leaves of water chestnut, *Trapa* sp., caused

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by larval and adult feeding of a leaf beetle, probably *Galerucella* sp. Almost all leaves were destroyed at one site. This beetle might be a potential candidate for control of water chestnut on Lake Champlain, New York.

Beijing

On 19 August I arrived in Beijing, where I met with Dr. Wang Ren and his staff at SABCL. Mr. Wang Yuan, who had helped with the project for 2 years, had left to take another position and had been replaced by Dr. Chen Ping-Ping. Dr. Chen Ping-Ping received her doctorate in Holland and is a world authority on the taxonomy of Saldidae, a family of semi-aquatic true bugs. Other staff who helped this year were Ms. Jiang Hua, who joined the project in 1990, and Ms. Liu Wei Zhen, an insect pathologist in SABCL.

During the first week, we sampled hydrilla and milfoil sites in Beijing for Dr. Balciunas and in order to obtain hydrilla leaf miners to initiate laboratory colonies that I could carry to Florida on my return trip. Both species of leaf-miner were common, *Hydrellia pакistanae* Deonier and *Hydrellia* n. sp. silver-face. We also sampled leaf-miners from associated plants, especially *Potamogeton* spp. and *Vallisneria* sp., to confirm the field host specificity of the hydrilla leaf-miners. New sampling sites this year included a small canal and a small lake at Hsing Hua University. Hydrilla and the leaf-miners were abundant in the canal, and milfoil was abundant in the lake. Unfortunately, when I returned to Beijing in early September, the hydrilla had been pulled from the canal and the heavy leaf-miner infestation that I had planned to carry to Florida had been lost.

The 1 August 1990 lake site still had milfoil with *Phytobius* sp. 1 weevils and scattered hydrilla with leaf-miners. Both weedy species coexist at that site with four or five other common American plant species or their close relatives, without dominating the site. Tip midge larvae were present on hydrilla at several sites again this year but were less

abundant than in 1990. This may have been because I was sampling earlier in the season this year than last. I am still enthusiastic about the damage caused by these midges and believe that we should study their field biology.

Xinjiang (Sinkiang) Province

On 25 August, Dr. Chen Ping-Ping, Mr. Fan (an administrative assistant at SABCL), and I flew to Urumqi (Ulamuchi) in the north-western province of Xinjiang. Our purpose was to survey for Eurasian watermilfoil insects near the Altay Mountains in the northern part of the province. According to herbarium specimens, milfoil was present in this area, and I hoped to find two species of weevils that I collected from milfoil in Kashmir, India, in 1985. Recent political instability has prevented travel in Kashmir, which is adjacent to southern Xinjiang. The *Bagous* larvae bore in milfoil stems, and the *Eubrychius* larvae and adults eat submersed leaves. I also hoped to find new species of insects because the area is far removed from our collecting locations in eastern China.

The area from Ulamuchi to the Altay Mountains was mostly desert except near waterways, where grass and trees were abundant. We traveled by car with our host, Mr. Jung, a young provincial official. Lakes, ponds, and small streams near the three cities of Altay (Altai), Fuhai (Fuhei), and Burqin (Buerjin) were searched in vain for Eurasian watermilfoil. Apparently another milfoil, *Myriophyllum verticillatum* L., had been misidentified as Eurasian watermilfoil, which might not be present in this area. This conclusion is supported by the fact that we found only *M. verticillatum* at a reported *M. spicatum* site and by the fact that Dr. Chen Ping-Ping observed misidentified herbarium specimens when we returned to Urumqi (Ulamuchi). Submersed leaves of the two species are quite similar, but the emersed flower stalk of *M. verticillatum* is much more robust with larger leaf-like bracts. No hydrilla was observed during the survey.

Near the Burqin River at Burqin we found two species of weevils attacking *M. verticillatum*. *Phytobius* sp. 2, which is larger than the species collected on *M. spicatum* in Beijing and elsewhere, attacked the emerged flower stalks. Larvae and adults appeared to feed on the bracts, and cocoons were formed in the submersed stems. Dr. Charles O'Brien, Florida A&M University, Tallahassee, believes that it is similar to but distinct from *P. leucogaster*, which is present on *M. spicatum* in Europe and North America. *Phytobius* sp. 2 was found at two sites near the city; at one site, almost all flower stalks had been stripped of bracts. Because larvae and adults were no longer present, we could not rule out the possibility that another agent, for example a caterpillar, was responsible. However, weevil cocoons were abundant without evidence of other insects.

Submersed leaves on *M. verticillatum* plants in a small slow-flowing stream near Burqin were eaten by both adults and larvae of *Eubrychius* sp., which is distinct from the species in Kashmir. Cocoons were formed in the stems close to the tips. Damage to the leaves appeared minimal, but the population was very small. This weevil swims well underwater and can remain submerged for long periods if the water is well aerated.

Adults of both species of weevils were carried to Beijing. The SABCL staff is attempting to rear *Phytobius* sp. 2, and I carried *Eubrychius* adults to Gainesville to determine if they would attack *M. spicatum*.

SABCL Trips

Dr. Chen Ping-Ping visited Chang Sha, Hunan Province, 22-27 September, to discuss the project with cooperators and to obtain their specimens and records. She also visited two lakes near Wuhan, Hubei Province, on 17 October. A *Phytobius* sp. was present on the flower stalks of *M. verticillatum* in Fu Tou Lake, and a *Hydrellia* sp. was present in hydrilla at Liang Zi Lake.

Quarantine Studies

Our return to Beijing from Urumqi (7 September) was delayed 4 days because of a flight mixup. Consequently, only 5 days remained for final sampling and preparation of a shipment of leaf-miners to carry to Florida. Unfortunately, the heavily infested canal site at Hsing Hua University had been manually cleared during our absence from Beijing, and I had to settle for a smaller shipment of larvae from our SABCL colony and from other sites. I departed Beijing on 13 September with a shipment of *Hydrellia* and *Eubrychius* sp.

In the Gainesville quarantine laboratory we obtained 54 females and 45 males of mixed *Hydrellia pakistanae* and *Hydrellia* n. sp. silver-face from this shipment. We did not separate the two species during the first two generations because males are not easy to speciate, and we wanted to maximize the males available to the females. However, we did separate the F3 generation into colonies of both species, and we are continuing to rear these flies apart from our colonies imported in 1990.

We have submitted a request to USDA/APHIS/PPQ for release from quarantine of the Chinese population of *H. pakistanae*. We are hoping that the request will be granted expeditiously without a delay of returning to the Federal Technical Advisory Group, which approved release of the Indian-Pakistani population of this species. Dr. Dick Deonier, Miami University, Oxford, OH, the original describer of *H. pakistanae*, has confirmed that the Chinese population is the same species. However, to err on the side of safety, we conducted cross-mating tests with the Chinese colony and the Pakistan colony, which at the time we had in greater number than the Indian colony. Reciprocal crosses were successful, and in the F2 generation we still had expanding populations.

We also conducted no-choice larval feeding tests with nine high-risk plant species

that had been tested during the original tests and two additional plant species. The results of these tests were essentially the same as the results of the original tests submitted in support of the request for field release of this species. Small numbers of adults were produced on several pondweeds and southern naiad, but none were produced when three of these species were tested in choice tests with hydrilla. We hope that these Chinese flies will be available next year for release at the northern hydrilla locations.

We are now in the second generation with the milfoil weevil, *Eubrychius* sp., however weevil numbers are very low. Fortunately, I had obtained a permit to import this genus prior to my 1990 trip to Kahsmir and China. It is unclear if these or the *Phytobius* weevils have potential for future utilization because of the type of damage they do and because there is already a similar native weevil fauna in North America. If we knew more about the importance of seed in milfoil populations, more about the potential invasion of pathogens through insect-feeding scars, and more about the damage to milfoil leaves by a native relative of *Eubrychius* sp., we might be able to better judge the potential of these Chinese species.

Most of our quarantine effort this year was directed toward clearing the Chinese *H. pakistanae* for release and in rearing and shipping the previously cleared weevils and flies, including the Australian stem-boring weevil

Bagous n. sp. Z. This weevil was released from quarantine to Dr. Center, who released it in the field. We also conducted some cross-mating and host range tests with the Indian and Chinese populations of *Hydrellia* n. sp. silver-face. Dr. Deonier has indicated that these populations appear to be one species that is similar to the Australian *H. balciunasi*. Cross-mating tests were successful one direction, but the reciprocal crosses were not. We need to continue tests to determine if this is a real difference.

Future Studies

During the next year we plan to ship the Chinese *H. pakistanae* to cooperators if we receive permission from APHIS. We also plan to continue host range and biological studies with the Chinese and Indian populations of *Hydrellia* n. sp. silver-face. If travel plans are approved, I hope to travel to Beijing and then to Harbin with SABCL staff to recover weevil larvae collected in hydrilla this year by Dr. Balciunas. Observations will be made on the field biologies of this species and of the milfoil weevil. My colleague Christine Bennett will travel to Beijing at the end of my stay and will remain to study the field biology of the tip midges and, if necessary, the field host range of *Hydrellia* n. sp. silver-face. We are hoping that Dr. Pemberton will be able to help with this project since Dr. Balciunas has stepped down to devote his time to the melaleuca project.

Release and Establishment of Insect Biocontrol Agents of Hydrilla in Alabama, Louisiana, and Texas

by

Michael J. Grodowitz¹ and Ed Snoddy²

Introduction

Hydrilla verticillata (L.f.) Royle is a submersed aquatic plant with a wide but rather disjointed geographical range (Pieterse 1981). In the United States, it is found throughout the southeastern states and recently has been causing problems in California. It appears to be spreading northward, with infestations established in northeastern Alabama and as far north as Delaware. Hydrilla causes manifold problems in many areas of its US distribution. Reasons for the development of such problems are related to hydrilla's growth characteristics, which include a relatively high growth rate, ability to grow in low light levels, and the production of two specialized organs (turions) used for surviving periods of stress (Pieterse 1981). These growth characteristics led to the development of large infestations of essentially monotypic hydrilla stands in the United States. Problems associated with such infestations include obstruction of navigation, restriction of water flow, hinderance of recreational uses, as well as impeded water use for the production of electricity.

Two broad technologies are currently used for the management of hydrilla. These include both mechanical (Haller and Joyce 1978) and chemical methods (Van Diver 1978). Unfortunately, both methods can be highly expensive and, typically, are not efficacious when hydrilla is present in large acreage. This has prompted several Federal and state agencies to search for viable alternatives for hydrilla management.

The use of insect biocontrol agents was identified as one alternative. While many researchers felt that such an approach would not be effective, limited information indicated that some species of insects were capable of producing significant damage on hydrilla. For example, Baloch, Sana-Ullah, and Ghani (1980) reported that a species in the family Ephydriidae (shore flies) was capable of producing considerable damage to hydrilla in Pakistan. This species, *Hydrellia pakistanae* Deonier, was subsequently brought into a US quarantine facility (Florida Biological Control Laboratory Quarantine Facility, Gainesville, FL) to begin biological and host range studies. It proved to be highly effective, and it was also restricted to feeding on hydrilla only (Buckingham, Okrah, and Thomas 1989). US field releases were subsequently made beginning in 1987. Similarly, a closely related Australian species, *Hydrellia balciunasi* Bock, was found to also produce considerable damage to hydrilla (Balciunas and Center 1988). *Hydrellia balciunasi* Bock was brought into US quarantine in early 1988, and field releases were made in the south Florida area beginning in 1989.

After the releases in the Florida area (see Center 1992), studies were initiated to increase the release and establishment efforts to other southeastern states. The following information will describe the initial release and establishment efforts for both *Hydrellia balciunasi* and *H. pakistanae* in Alabama, Louisiana, and Texas. It is important to remember that these release and establishment

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efforts are a product of a cooperative program involving the US Army Corps of Engineers (USACE), two USACE Districts (the New Orleans District and Galveston Districts), the Tennessee Valley Authority (TVA), and the State of Texas Parks and Wildlife Department.

Materials and Methods

Site locations

As indicated previously, three states in addition to Florida have had releases of either *H. balciunasi* or *H. pakistanae*. In Alabama, *H. pakistanae* has been released at three locations, including two sites on Lake Guntersville and one general area in the Muscle Shoals vicinity. *Hydrellia pakistanae* has also been released in Louisiana on Lake Boeuf, a small marshy lake near the town of Raceland. In the state of Texas, *H. balciunasi* was released on Sheldon Reservoir, located on the northeastern side of Houston. *Hydrellia balciunasi* was released only in Texas for two reasons: we wanted to evaluate the impact of *H. balciunasi* without the influence of *H. pakistanae*, and *H. balciunasi* is originally from an area of Australia that is similar climatically to the Houston area.

Insect sources and release techniques

All released *Hydrellia* were obtained from rearing facilities located at either the US Army Engineer Waterways Experiment Station (WES) or a TVA facility located in Muscle Shoals. Rearing procedures were similar to those described by Buckingham, Okrah, and Thomas (1989). The current rearing procedures lend themselves well to mass-rearing techniques. For example, the WES rearing facility has released >300,000 *Hydrellia* since its inception in 1990.

Both *H. pakistanae* and *H. balciunasi* were released from the various rearing facilities as late 2nd or 3rd instar larvae contained within infested plant material. The infested plant material was transported to the release site wrapped in water-soaked paper toweling.

The infested plant material was then carefully placed within a 1-m² floating cage. The cage was designed to limit the initial dispersal of the emerging adults. The cage also ensured that the limited number of emerging adults would remain in the general release area and that large enough numbers would be in contact to ensure mating and subsequent oviposition in the area.

Insect and plant monitoring

An essential part of any release and establishment program is both a qualitative and quantitative monitoring program to verify establishment, subsequent plant damage, and insect population levels. This information is needed to verify successful establishment of the released insect populations and serves as a guide for future release efforts.

Qualitative sampling

Qualitative sampling for *Hydrellia* is the primary means of assessing establishment. Qualitative sampling allows for a rapid assessment of insect establishment at several locations across wide geographical ranges with limited disturbance to the release area. Sample collection can be accomplished by personnel not specifically trained in insect biocontrol research. Essentially, qualitative sampling allows for the collection of at least some information on the status of the hydrilla, verification of *Hydrellia* establishment, as well as insect damage.

Qualitative sampling was accomplished by collecting large numbers of adult *Hydrellia* from the area surrounding the release site and by collecting nearby rooted hydrilla to assess larval damage and subsequent adult emergence. Adults were collected by moving a large sheet of styrofoam in front of a moving boat. The adult *Hydrellia* resting on the water and plant surface were disturbed by this activity, and the adults tended to land, in large numbers, on the floating styrofoam. The adults were then collected from the sheet by carefully placing a small vial over the resting adult.

This collection technique not only gathers *Hydrellia pakistanae* and *H. balciunasi* but also several species of native *Hydrellia*. Identification was made using characteristics supplied by Dr. Dick Deonier, including abdominal length in relationship to thorax length as well as genitalia characters. Continued presence of adult *Hydrellia* in the general release area is evidence of successful establishment. The presence of adult *Hydrellia* is especially important information if *Hydrellia* releases had been discontinued in the release area.

Two qualitative measurements are performed on rooted hydrilla collected from areas adjacent to the release area. The rooted hydrilla material is used primarily for collection of emerging adults. The collected hydrilla is placed into 2-L clear plastic containers filled with fresh lake water or deionized water. The containers are held under greenhouse conditions under a constant temperature of approximately 20 °C. Temperatures are held constant via the use of waterbaths. The containers are checked daily for emerging adults. The adults are collected and subsequently identified.

In addition, small quantities of the collected hydrilla are examined microscopically for the presence or absence of the immature *Hydrellia*. In some instances the collected hydrilla is placed within a Berlese funnel, a collecting device that slowly dries the plant material, allowing the entrapped insects to migrate away from the drying source (i.e., light bulb) to a collecting vial filled with 70 percent ethanol. The resulting alcohol and insect mixture is subsequently examined for both *Hydrellia* larvae and adults.

While these techniques do not specifically measure plant and insect status quantitatively, they do serve as overall indicators for establishment, plant damage, insect presence, insect numbers, and plant population status. Evidence of the presence of *Hydrellia* over a continued time interval is strong indication of successful establishment.

Quantitative sampling

While qualitative sampling provides strong evidence for successful establishment, it does not provide enough information to actually confirm establishment or indicate the degree of impact caused by the insect agents. This information is collected by routinely sampling, quantitatively, both the insect and plant population until a picture of the population dynamics is obtained. Changes in plant dynamics in relation to insect population fluctuations are a strong indication of a biocontrol effect. While quantitative sampling is time consuming and expensive, it is one of the best ways to provide enough information to confirm establishment and indicate that the observed impact is caused by the released insects and not by other environmental factors. In addition, quantitative sampling supplies much-needed information on the mechanisms concerning insect damage and associated impact, the relationships between insect numbers and plant damage, and the best method for future plant and insect sampling and releases.

Quantitative sampling for plant population dynamics was accomplished by determining plant biomass per unit area. Quantitative sampling was accomplished at the Lake Guntersville site only. At this site, biomass was partitioned into that portion of the hydrilla mat forming a canopy at the water surface (i.e., approximately 15 to 30 cm below the water surface) and the remaining plant material to the hydrosoil (i.e., bottom).

Ten randomly selected 10- to 20-cm-long hydrilla pieces from each partition were weighed and the number of whorls quantified. The leaves were subsequently removed from these stem pieces and the leaf and stem weights determined. Dry weights were determined by collecting subsamples from each plant portion, as well as the leaf and stem pieces, and drying at 60 ± 2 °C.

Insect numbers and their damage were determined by randomly collecting ten 10- to

20-cm-long stem pieces and carefully examining for the presence of larvae and pupae. In addition, the number of leaves was counted and the distribution of damaged leaves quantified. Distribution classes included leaves <25 percent damaged, 25 to 50 percent damaged, and >50 percent damaged. Number of insects per unit biomass was determined by the total weight examined for insect larvae and pupae in comparison to total plant biomass collected.

Plant samples were collected using a submersible frame device (modification of design by Sallie P. Sheldon). The sampling device consisted of a 0.25-m² frame approximately 5 ft in height enclosed with either Plexiglas or plastic sheeting. The frame was carefully lowered into the hydrilla mat by scuba divers. The divers ensured that the hydrilla enclosed by the frame stayed within the frame boundaries and did not become entangled by the sides of the sampling device. After total immersion into the mat (i.e., to the hydrosoil), the hydrilla was clipped at both the sediment and the canopy levels. Each section was separated by a tight-fitting mesh and brought to the surface. The mesh ensured that *Hydrellia* larvae would not be lost by water runoff from the collected plant material.

Results and Discussion

Alabama

As indicated previously, releases have been made in Alabama at three distinct locations. These have included one site on Lake Guntersville (including two general areas approximately 1 mile apart—Chisenhall and Comer Bridge); the Murphy Hill Aquatic Plant Research ponds in Guntersville, AL; and the Muscle Shoals, AL, pond facility. The Lake Guntersville sites were chosen to determine if establishment of *H. pakistanae* can be accomplished on a large reservoir and whether, when successful establishment occurs, the insects can survive the winter season. If establishment and overwintering are successful, impact can be assessed adequately.

On the other hand, the pond facilities were to serve as nursery areas for future releases as well as to verify establishment and overwintering capacity under more controlled conditions.

Beginning in late August 1990 and continuing through the end of October 1990, a total of 29,512 late 2nd instar and 3rd instar *H. pakistanae* larvae were released in the Comer bridge area on Lake Guntersville (Table 1). Adults were collected within the release cage during September 1990, indicating that at least some of the released larvae were surviving to the adult stage. Limited qualitative sampling in the area adjacent to the release cage did not indicate the presence of any life stage of *H. pakistanae*.

During September and October 1990, significant changes occurred to the hydrilla near the original release site as well as throughout the hydrilla infestation from the Chisenhall to Comer Bridge area. A majority of the hydrilla canopy disappeared, and the only remaining hydrilla formed a thick mat or "carpet" near the sediment surface. Grass carp had been released by TVA as part of their aquatic plant management strategy and were cited as the probable cause for the plant disappearance. Grass carp were observed feeding in the vicinity during this time.

Because of the presence of grass carp, two large exclosures (approximately 1,000 m²) were constructed during May 1991 at the original release area (Comer Bridge) and approximately 2.5 km downstream (Chisenhall). The exclosures were constructed from fishery block nets and were designed to exclude grass carp from the test areas. To ensure that grass carp were not confined within the exclosures during construction, electroshocking boats were operated within the exclosures before they were sealed.

Differences in plant composition were observed by June 1991 at the two sites. While hydrilla was the predominant plant species in the Chisenhall exclosure (approximately 95 percent), only limited quantities of hydrilla

were observed in the Comer bridge enclosure (approximately 30 percent). The majority of the remaining plant material was Eurasian watermilfoil. To reduce the quantity of milfoil, 2,4-D was applied to both exclosures. However, by July 1991 it was evident that hydrilla was to remain at minimal levels at the original *H. pakistanae* release area (Comer Bridge). This prompted us to shift our insect release area to the Chisenhall enclosure.

Beginning on July 11, 1991, and ending August 8, 1991, a total of >25,000 late 2nd instar and 3rd instar larvae were released in the Chisenhall enclosure (Table 1). During July and August 1991, several qualitative measurements indicated that *H. pakistanae* was becoming established at the Chisenhall area. Adult *H. pakistanae* were collected during hand-collection efforts as well as from floating soap dishes that trapped the adults. In addition, larvae were collected from nearby rooted hydrilla, indicating successful transfer

of the larvae from the infested sprigs. Larvae were also collected from rooted hydrilla after releases were terminated in early August 1991. Only one quantitative sample collected during August contained immatures. From this sample we estimated that about 1 pupae was present per m², and only a limited number of damaged leaves was observed. No *H. pakistanae* were collected at the Comer Bridge enclosure; in fact, only limited hydrilla was collected throughout the growing season at this area (Figures 1 and 2).

Soon after releases were terminated in August, significant changes were observed in the hydrilla status at the Chisenhall enclosure. For example, a 40-percent reduction was observed in the total dry biomass collected from this area from the August to the September 1991 sampling effort (Figure 1). Apparently, large losses occurred in the canopy as indicated by minimal canopy growth during July and a complete loss by September 1991 (Figure 2). Growth appeared to continue in the bottom portion, as indicated by increases in total mean dry weight until September 1991 followed by small reductions of approximately 7 percent in October 1991. Dry leaf and stem weights for the bottom portion also increased substantially throughout the sampling period in the Chisenhall enclosure (Figure 3). Little hydrilla growth was observed at the Comer Bridge enclosure (Figure 3).

Explanations for the rapid loss of hydrilla from the Chisenhall enclosure are not available. Grass carp could have entered the enclosure, but no evidence of the fish was found, even after extensive electroshocking within the enclosure during September 1991 and numerous visual observations throughout the periods of loss.

Examination of the plant material from the Chisenhall enclosure after the canopy loss during the September sampling indicated the presence of distinct brown areas on a majority of the leaf tips. Isolation from this plant material revealed the presence of several potential fungal pathogens. The extent of loss caused by their presence is unknown.

Table 1
Releases of *Hydrellia pakistanae* on Lake Guntersville, August 1990-August 1991

Date	Number Released
08/28/90	1,226
09/04/90	3,235
09/06/90	2,053
09/13/90	2,352
09/20/90	1,642
09/26/90	3,055
10/03/90	2,306
10/09/90	2,236
10/10/90	3,455
10/22/90	1,465
10/24/90	2,550
10/30/90	3,931
Total	29,506
Date	Number Released
07/11/91	20,289
07/22/91	6,921
08/05/91	1,984
Total	29,194

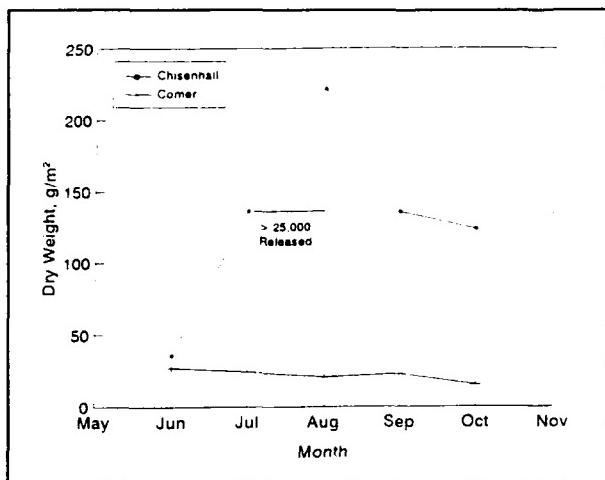


Figure 1. Total dry weight of hydrilla collected from Chisenhall and Comer Bridge areas on Lake Guntersville, June 1991 to October 1991

Another plausible reason for the disappearance of hydrilla is a potential interaction between *H. pakistanae* and the pathogens. It is possible that the feeding damage caused by *H. pakistanae* may have allowed the entrance of a pathogen that otherwise would have remained innocuous. In addition, the stress placed upon the hydrilla by the insect feeding may have allowed an otherwise innocuous pathogen to become damaging. We are now in the process of evaluating the combined effects of the pathogen strains found at Chisenhall and *Hydrellia pakistanae* on hydrilla status under more controlled experimentation.

Releases have also been made on small ponds located at both Guntersville and Muscle Shoals, AL. A total of 9,670 2nd and 3rd instar *H. pakistanae* were released at the Murphy Hill pond. These releases were made during August and September 1991. Qualitative sampling has indicated that establishment was occurring since both adults and larvae were found in areas adjacent to the original release area. No changes in the hydrilla status have been observed.

More than 12,633 2nd and 3rd instar *H. pakistanae* were released on one pond in the Muscle Shoals area during August and September 1991. Similarly, establishment is believed to be occurring, since adult and larvae were collected in areas adjacent to the original

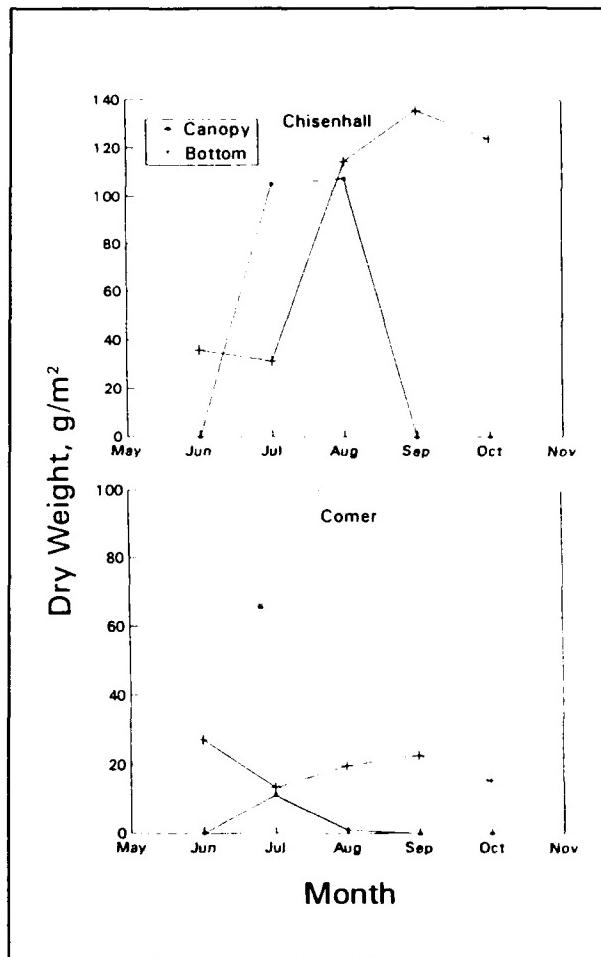


Figure 2. Total dry weight shown in Figure 1 partitioned into that plant material forming mat near water surface (canopy) and the remaining plant material to the hydrosoil (bottom) collected from Chisenhall and Comer Bridge areas on Lake Guntersville, June 1991 to October 1991

release sites based on repeated qualitative sampling. Again, no distinct changes have been observed in the hydrilla status.

Louisiana

Working cooperatively with personnel of the New Orleans District, releases of *H. pakistanae* were begun on Lake Boeuf beginning in July 1991 (Table 2). More than 76,000 2nd and 3rd instar *H. pakistanae* were released over a 3-month period. The number of insects per release averaged 6.000. As with the releases in Alabama, qualitative sampling provided evidence that establishment

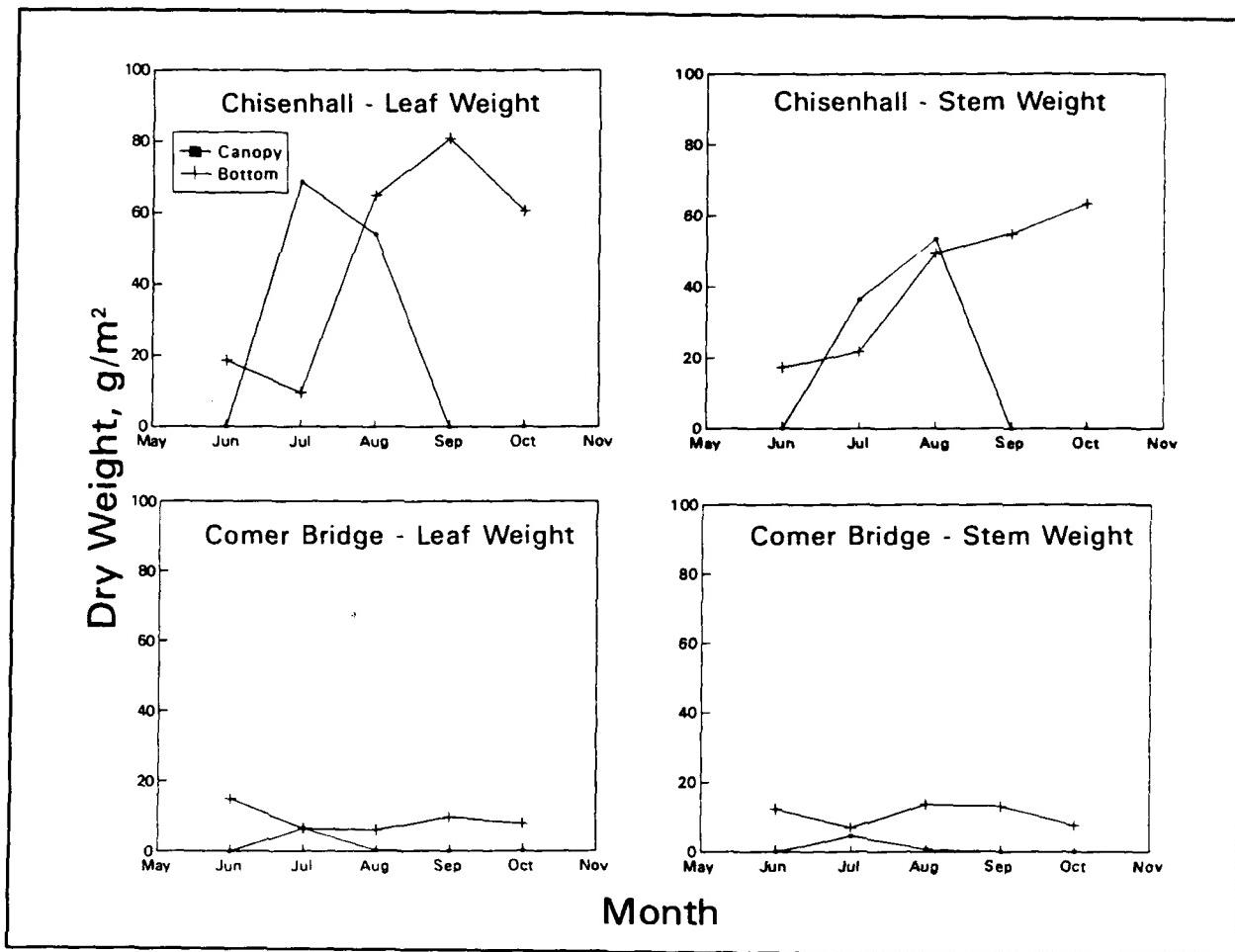


Figure 3. Dry weight of leaves and stems from the canopy and bottom portion of hydrilla collected from Chisenhall and Comer Bridge areas on Lake Guntersville, June 1991 to October 1991

Table 2
Releases of *Hydrellia pakistanae* on Lake Boeuf, July 1991–October 1991

Date	Number Released
07/24/91	9,322
07/30/91	11,943
08/14/91	11,259
08/20/91	7,111
08/27/91	2,459
09/03/91	3,714
09/10/91	4,976
09/17/91	3,784
09/24/91	1,778
10/10/91	3,007
10/17/91	5,563
10/21/91	4,614
10/28/91	6,749
Total	76,279

was occurring. Both adults and larvae were collected as far away as 1 m from the original release area.

As with the Guntersville reservoir site, distinct changes were observed in hydrilla status during August and September 1991. Massive losses of the hydrilla canopy were noted immediately adjacent to the original release site. The affected area, where only minimal hydrilla was found, exceeded 1.5 acres. Examination of the remaining hydrilla revealed that many of the leaves were totally missing, with only denuded stem pieces remaining. Pathological isolations did not reveal the presence of any potential pathogens.

After the hydrilla loss, the release area was moved to a site approximately 1 km

from the original release area. The site movement occurred during early September 1991. Only minor changes have been observed in the hydrilla status in this area, and these may be the result of seasonal changes and not insect feeding.

Texas

Hydrellia balciunasi was released at one site in Texas during the 1991 growing season. This site was a small pond area adjacent to Sheldon Reservoir located on the northeastern side of Houston, TX. It was selected because of its limited public access, because information on the hydrilla infestation was available from other biocontrol studies that had been conducted in the area, and because the climatic conditions were similar to the home range of *H. balciunasi*. Approximately 20,000 2nd and 3rd instar *H. balciunasi* were released beginning in August 1991 and continuing through November 1991.

As with sites in Alabama and Louisiana, repeated qualitative sampling has indicated that establishment is occurring. As late as October 1991, adult *H. balciunasi* have been collected from the area adjacent to the original release site on Sheldon Reservoir. However, no changes have been observed in the hydrilla status.

Future Studies

The major thrust for the 1992 growing season is to expand our monitoring at each of the previously mentioned release areas. Both qualitative and quantitative sampling will continue at an increased frequency. For example, on Lake Guntersville, quantitative sampling will be accomplished bimonthly instead of monthly as in 1991. Both qualitative and quantitative sampling will be accomplished on Lake Boeuf, and weekly qualitative samples will be taken at the Sheldon Reservoir site.

After we have completed the initial qualitative sampling efforts to determine overwintering survival, decisions concerning future

releases will be made. We hope to continue our release program in the present locations as well as increase the number of release locations in each state. In addition, we will examine the possibility of releasing other hydrilla biocontrol agents in these states. This would include the release of the cold-tolerant *H. pakistanae* from Beijing, China, recently released from quarantine in the Lake Guntersville area. We will also entertain the idea of releasing the stem-feeding hydrilla weevil, *Bagoous* new species.

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The Impact of Temperature on *Hydrellia pakistanae*

by

Ramona H. Warren¹

Introduction

Hydrilla verticillata (L.f.) L. C. is a submersed aquatic plant with a high reproductive potential and wide ecological tolerance (Baloch and Sana-Ullah 1973). The plant is located throughout the southern United States, along the east coast as far north as Delaware, and in southern California (Cofrancesco 1991). This nuisance aquatic plant causes problems by interfering with water flow and recreational activities in lakes and rivers. It also impedes navigation by clogging waterways.

The methods used to control hydrilla are mechanical, chemical, and biocontrol. Mechanical control is accomplished by using machinery such as mechanical harvesters. Chemical control is accomplished by the use of herbicides. Biocontrol control is accomplished by using natural enemies to manage exotic aquatic plants.

In large areas of plant infestations, mechanical and chemical methods of control may not be as effective or efficient as other control methods (Hamilton 1991). This prompted researchers to seek other methods of control. The study of biocontrol technology for aquatic plant management began in 1959 when the US Army Corps of Engineers and the US Department of Agriculture (USDA) entered into a cooperative study of this control method (Cofrancesco 1991). As a result of this study, the *Hydrellia pakistanae*, an ephydrid fly from Pakistan, was released in the United States (1987 in Florida) as a biocontrol agent for hydrilla.

The impact of temperature on the development of *H. pakistanae* is important in deter-

mining if insect populations can be established in all regions in which hydrilla is located. This study examines the impact of a sample set of temperatures on *Hydrellia pakistanae* developmental stages from egg to adult.

Methods and Materials

Plants and insects

The experiment was initiated by obtaining insect eggs from the Waterways Experiment Station (WES) colony of *Hydrellia pakistanae*. The plant material (hydrilla) was obtained from the WES greenhouse facilities.

Experimental setup

Nine 15-cm sprigs of hydrilla were placed in a petri dish and put into the colony chamber for oviposition for a 24-hr time period. The eggs were then retrieved and placed in petri dishes. Each petri dish contained a sheet of black filter paper and 20 individual leaves. A single egg was placed on each leaf. These petri dishes were then placed into environmental chambers at the desired test temperatures (20, 25, and 27 °C) with a 14-hr photophase. The petri dishes were checked daily for egg hatch. First instar larvae were placed into fifty 60-ml test tubes (one larva per tube). Each tube was filled with deionized water and contained a 15-cm sprig of hydrilla. Each hydrilla sprig had 15 to 20 whorls of four or five leaves. The tubes were covered with nylon organdy and held in place with a rubber band. (Note: The tubes were placed in an environmental chamber at the same time as the five petri dishes so that the environment would be the same for the insect when transferred to the test tube.)

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Observation

The test tubes were examined daily, for the duration of each study, under a dissecting microscope for (a) development of life stages, (b) location of the insect on the plant, (c) percent leaf damage of the hydrilla leaves, and (d) percent survival.

Data analysis

The data were analyzed using the Mean Separation Technique of the Statistical Analysis System (SAS), a computer statistical software program. This program produced the means and standard error of the data collected.

Results and Discussion

Figure 1 shows a graphical chart of the average developmental rate at each life stage for all three test temperatures. Note from this

chart that shorter developmental rates were associated with higher temperatures. The 25 and 27 °C life stages from egg to adult were not noticeably different when compared to each other, but when these two stages were compared to the 20 °C life stages, they were approximately twofold less than the 20 °C life stages from egg to adult. Also note that the differences were most apparent in the egg and pupa stages for all three test temperatures.

The development rates from egg to adult at temperatures of 20, 25, and 27 °C were 43, 26, and 23 days, respectively. These rates were similar to the results of earlier study on the development rate of *H. pakistanae* (Buckingham and Okrah, in press). In this study, the development time period was the same at 27 °C.

Table 1 shows the percent survival for *H. pakistanae* life stages. From this table note

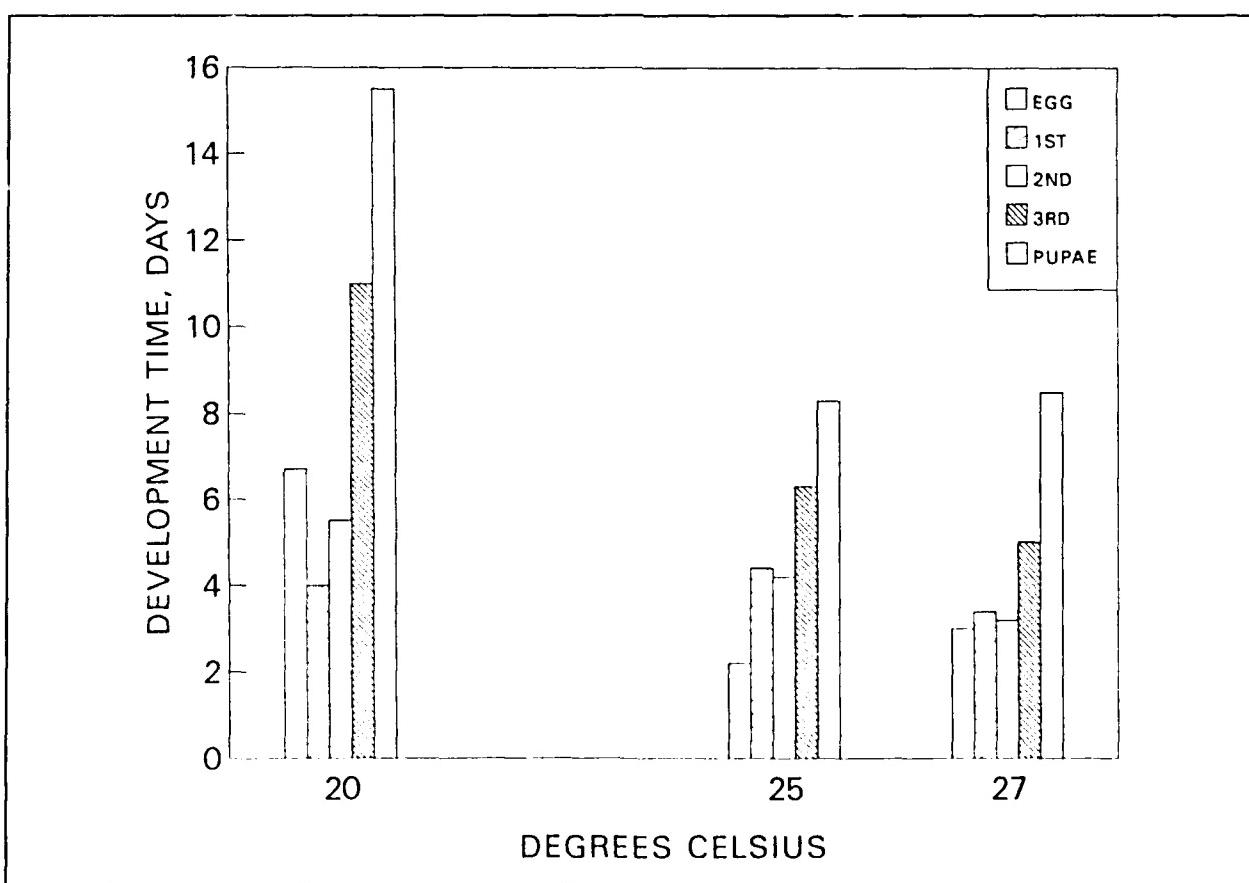


Figure 1. Average development rate at each life stage for all three test temperatures

that the best temperature for survival appeared to be 25 °C, which was 36 percent higher than the temperature at 20 °C and 32 percent higher than the temperature at 27 °C. In fact, 25 °C appeared to have resulted in a higher survival rate in all larvae stages.

Table 1
Percent Survival for *Hydrellia pakistanae* Life Stages

Temper- ature, °C	% Egg	% 1st Instar	% 2nd Instar	% 3rd Instar	% Pupa
20	100	78	68	58	54
25	100	94	90	86	84
27	100	81	65	57	57

Table 2 shows the average number of leaves damaged by the larvae in the experiment. From this table, note that the damage due to mining during the larval stages exhibited minor differences with respect to temperature in the first and second instar, but in the third instar, the damage at 20 °C is approximately twofold higher than at 25 and 27 °C.

Table 2
Average Number of Leaves Damaged by Larval Stage

Temperature, °C	Total	1st Instar	2nd Instar	3rd Instar
20	10.84	1.39	2.73	7.08
25	8.20	1.26	2.72	4.30
27	5.72	1.12	1.60	3.04

Conclusions and Future Research

Based on findings presented, of the three test temperatures, the development rate at 25 °C is close to the development rate at 27 °C but resulted in higher survival than at both 27 and 20 °C. These two factors indicate this temperature to be the best rate of the three test temperatures.

Present research will continue on developmental stages as they relate to temperature. Cold-temperature studies will also be conducted. From these studies, high and low threshold temperature limits will be determined for *H. pakistanae* developmental life stages.

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Waterlettuce Biocontrol Agents: Releases of *Namangana pectinicornis* and Dispersal by *Neohydronomus affinis*

by

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Introduction

Waterlettuce (*Pistia stratiotes* L.) is a floating macrophyte that impedes irrigation practices, hampers flood control efforts, blocks navigational channels, and interferes with recreational uses of many waterways in the United States. A biological control program against this nuisance was initiated in 1985 when *P. stratiotes* populations in Florida were surveyed for native herbivores (Dray et al. 1988). Quarantine studies later resulted in approval to release two foreign biocontrol agents (Habeck et al. 1989; Thompson and Habeck 1988, 1989). The first, a weevil (*Neohydronomus affinis* Hustache), had previously been applied successfully against waterlettuce in Australia (Harley et al. 1984) and later Africa (Cilliers 1987, 1990). The second, a moth (*Namangana pectinicornis* Hampson), is used to prevent destruction of rice paddies in Thailand by this plant (Napompeth 1982). It has also been recommended for use in the Philippines (Bua-ngam 1974) and Indonesia (Mangoendihardjo 1983).

Field releases of *N. affinis* began in spring 1987, and persistent populations had become established at several sites by fall 1988 (Habeck et al. 1989, Dray et al. 1990). Although weevil populations remained only marginal at some sites (e.g. Port St. Lucie), the insects clearly prospered at others. By the end of spring 1989, Kreamer Island on Lake Okeechobee harbored approximately 45 million *N. affinis* (Center and Dray 1990). The following year (1990), weevil abundance

at Torry Island on Lake Okeechobee reached similar proportions. Waterlettuce populations at both sites declined to less than 10 percent of preweevil densities.

Failure by *N. affinis* to persist at some sites or to build to densities that stress waterlettuce at others strengthened perceptions that the program could benefit from the release of a second biocontrol agent. Thus, *N. pectinicornis* was added to the arsenal of insect herbivores being applied against waterlettuce (Habeck et al. 1989, Center and Dray 1990, Grodowitz 1991). Field releases began in December 1990, but establishing persistent field populations has proven elusive.

This paper documents our efforts, thus far, to establish *N. pectinicornis* on waterlettuce in Florida. Further, it discusses continued dispersal by *N. affinis* and outlines our future plans regarding these two waterlettuce bioagents.

Culture Techniques

The techniques we employ to culture *N. pectinicornis* are modifications of the methods developed by Dr. Dale Habeck and his assistants during the quarantine testing of this moth in Gainesville. Most commonly, we introduce adult moths into $\frac{1}{2}$ " x 1" boxes containing whole plants and allow them to oviposit for one to several days. Leaves containing egg masses are then excised from the plants and placed on paper towels (moistened with benomyl, a fungicide) in petri dishes. Eggs

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are monitored daily, and following enclosion, larvae are transferred to fresh leaves and placed in large, clear plastic containers. Leaves are replaced as needed. Pupae are removed from these containers and placed on moist filter pads in petri dishes. Adults that emerge are introduced into a glove box to continue the cycle.

In a variation of the preceding technique, we introduce a pair of moths into a petri dish containing one or two leaves on benomyl-moistened filter paper. Leaves upon which eggs have been deposited are then handled as described previously. This technique is as effective as the earlier method and is preferable for obtaining accurate assessments of oviposition and larval survival. However, it is more labor intensive.

To reduce the labor involved in culturing this insect, we have also released adults into screened outdoor tanks (400-gal capacity) or greenhouse tanks (800-gal capacity) containing waterlettuce. Several generations of moths are allowed to complete development without being disturbed. Then, infested plants are removed to inoculate new tanks, or for release at field sites. Fresh plant material is introduced into the tanks as needed.

Colonies thus developed were very productive when laboratory-bred plants were used as the food source. However, there seemed to be a general reduction in productivity when plants from the Eagle Bay site were used. This needs to be tested more formally, but suggests that waterlettuce from Eagle Bay may be nutritionally deficient relative to laboratory-bred plants. Alternatively, *Samea multiplicalis*, a native waterlettuce herbivore that infested plants from Eagle Bay, may out-compete *N. pectinicornis*.

In another technique, we remove all but four leaves from a waterlettuce ramet. Each remaining leaf is enclosed in a sleeve of fine-meshed, nylon netting that is secured around the base of the leaf. A pair of adult moths is introduced into the sleeve and allowed to oviposit. After the adults die, leaves containing

egg masses are removed and placed into containers. These are then processed in the manner described previously. This technique yields surprisingly few viable egg masses, despite the close similarity to method 1 described above. Perhaps females are more likely to mate in the presence of several males rather than a single male, or perhaps the cage does not provide adequate space for mating rituals. Alternatively, the excision of some leaves may have released volatile compounds from the plant that served to deter females either from mating or ovipositing.

A final technique we have tested is a modification of methods previously developed (by T. Center) to obtain *Arzama densa* eggs. In this method, adult moths are introduced into cages constructed of large-mesh plastic screening. In the absence of plant material, we expect the moths to oviposit on wax paper that lines the exterior of the cages. Areas containing egg masses are then clipped from the wax paper and placed in petri dishes. Enclosing larvae are removed, placed on fresh leaves in plastic containers, and handled as previously described. This technique proved very inefficient for obtaining viable *N. pectinicornis* eggs.

Several of the techniques we have examined are capable of producing large numbers of *N. pectinicornis*. We will continue to employ these techniques even as we test additional methods for increasing the productivity of our laboratory colonies, both through attempts to enhance oviposition success and through efforts to improve larvae survival.

Project Status

Namangana pectinicornis

Nearly 110,000 *N. pectinicornis* eggs, larvae, and adults have been released at 11 field sites in Florida (Table 1, Figure 1). This includes 58,000 insects from the Fort Lauderdale laboratory and 52,000 insects from colonies maintained in Gainesville. The majority of the insects were released at four sites: Fish-eating Creek, Eagle Bay (Lake Okeechobee),

Table 1
***Namangana pectinicornis* Release Sites**

Site	County	Releases		No. Released			Plants ¹
		No.	Date	Eggs and Larvae	Pupae	Adults	
Fisheating Creek ²	Glades	13	12/18/90-4/22/91	22,617	0	0	No
Eagle Bay (L. Okeechobee) ²	Okeechobee	12	4/24/91-8/12/91	31,163	30	100	Yes
Lake Oklawaha ²	Putnam	10	2/13/91-8/30/91	28,100	15	4	No
South Florida Fairgrounds ³	Palm Beach	1	12/5/90	1,500	0	0	No
Pioneer Park ³	Palm Beach	1	12/30/90	2,347	0	0	No
Port St. Lucie ³	St. Lucie	2	12/1/90-12/30/90	3,208	0	0	No
St. John's Marsh ³	Brevard	2	1/2/91-4/25/91	4,468	0	0	No
Loxahatchee Recreation Area ³	Broward	1	9/9/91	3,000	0	0	No
Havana Pond ³	Gadsden	2	9/7/91-9/11/91	1,500	0	0	No
Lake Panasofkee ³	Sumter	2	9/25/91-9/30/91	1,200	0	0	No
Andytown ⁴	Broward	24	11/6/91-12/31/91	23,352 ⁵	127	101	Yes

¹ Whole waterlettuce plants (as opposed to individual leaves) infested with unknown numbers of *N. pectinicornis* larvae and eggs.
² Sites where we made multiple releases of large numbers of *N. pectinicornis*.
³ Sites where we made few releases of small numbers of *N. pectinicornis*.
⁴ Site where we are currently making intensive releases of *N. pectinicornis*.
⁵ This figure includes estimates of number of eggs contained in egg masses.

Lake Oklawaha (Rodman Reservoir), and Andytown (a canal bordering U.S. 27 north of S.R. 84). Additional release sites (Table 1, Figure 1) include a borrow pit near the South Florida State Fairgrounds, a canal bordering Pioneer Park in Belle Glade, a canal in Port St. Lucie, St. John's Marsh near Palm Bay, Loxahatchee Recreation Area, Havana Pond, and Lake Panasofkee.

Three progressively more intensive release strategies have been employed in attempting to establish *N. pectinicornis* populations at field sites. We originally thought that establishing field populations of this moth would be a simple matter of releasing a few insects at any given site, then allowing numbers to build up naturally. We therefore released small numbers of eggs and larvae at several field sites in southern Florida (see Table 1). Preliminary examinations 1 month after release at the Port St. Lucie site were encouraging. *Namangana pectinicornis* larvae were recovered from waterlettuce about 100 m from the release point. Unfortunately, later visits

failed to produce additional evidence that *N. pectinicornis* populations had established.

The results at Port St. Lucie, and from other sites, convinced us that establishing *N. pectinicornis* populations at field sites would be more difficult than originally envisioned. Consequently, we began employing a strategy of multiple releases comprising large numbers of insects at few sites (Fisheating Creek, Eagle Bay, and Lake Oklawaha; see also Table 1). Preliminary evidence suggested we were close to having established *N. pectinicornis* at Fisheating Creek when floods flushed the release site clean of plants.

Subsequent efforts at Eagle Bay were unsuccessful despite the release of nearly 32,000 *N. pectinicornis* there. Some of these were released into cages for protection from predators and to improve mating success by constraining the dispersal of emerging adults. Employing these extra precautions, which have been successfully employed with hydrilla flies (Center, Dray, and Durden 1991),

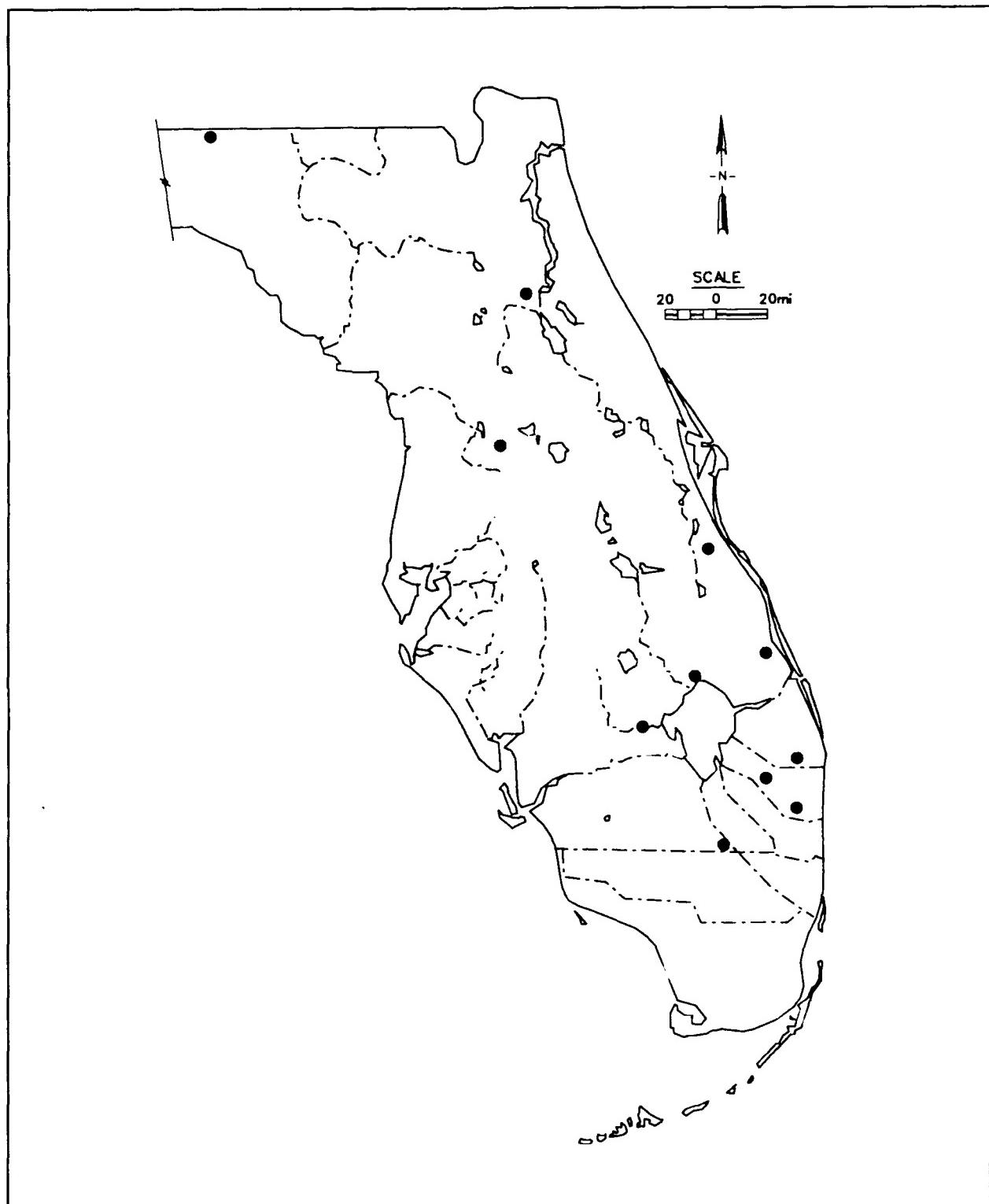


Figure 1. *Namangana pectinicornis* release sites, December 1990-December 1991

seemed ineffective with this bioagent at this site, however.

The failure of these two release strategies has caused us to rethink our approach. Consequently, we have initiated a highly intensive strategy in which we release large numbers of *N. pectinicornis* several times a week at a single nearby site (Andytown, see Table 1). Close monitoring of the site is providing much-needed feedback concerning the short-term fate of released individuals. Thus far, we have observed larvae dispersing from the leaves on which they were released to plants at the site. Although it would be premature to draw definitive conclusions, especially in light of earlier disappointments, the preliminary results are encouraging.

Neohydronomus affinis

We have observed control of waterlettuce by *N. affinis* at five sites: Kreamer Island (Lake Okeechobee, Palm Beach County), Torry Island (Lake Okeechobee, Palm Beach County), Plantation Golf Club pond (Broward County), and two Tenoroc State Preserve ponds (Polk County). Weevils continue to disperse from these sites to infest adjacent water bodies. At least 45 Florida waterways, only one third of which were release sites (Figure 2), have become infested *N. affinis* since the first populations were established on Lake Okeechobee in 1988. Further, Grodowitz (1991) reported that *N. affinis* populations have become established at six sites in Louisiana, from which they are also presumably dispersing.

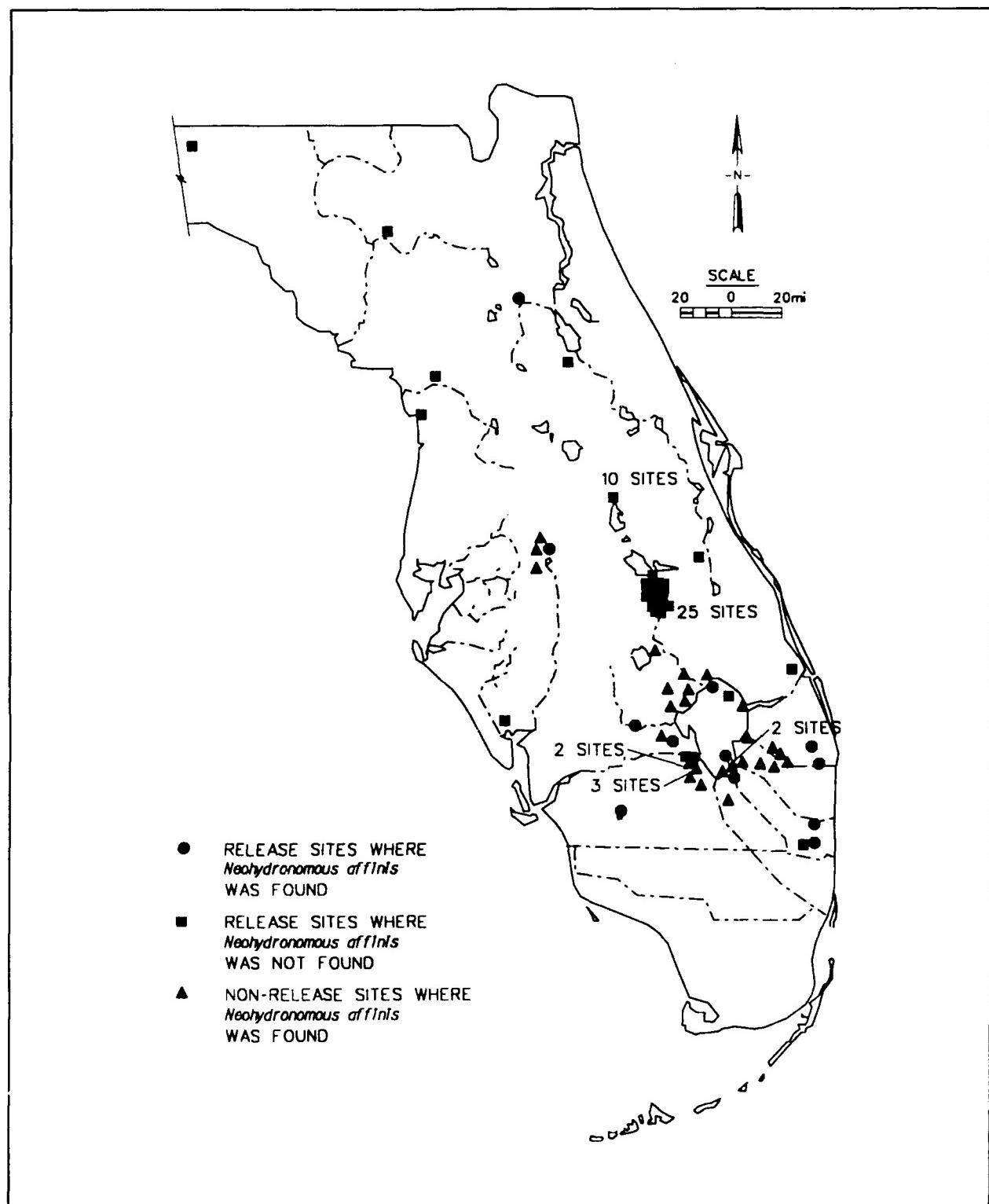
We anticipate that the weevil will continue to expand its range without human intervention, but it seems to be doing so rather slowly. Therefore, we are working closely with cooperators (both operational and research personnel) from Federal and state agencies to transfer insects from infested to

noninfested waterlettuce sites. For example, *N. affinis* were transferred from Tenoroc State Preserve to Lake Oklawaha (Rodman Reservoir) and Lake Rousseau (Levy and Citrus Counties) during August 1991. Unfortunately, the waterlettuce population at Lake Rousseau was treated with herbicides a short time after the weevils were released. Recovery of *N. affinis* larvae at Lake Oklawaha in November (8 weeks after release of infested plants), while not conclusive regarding population establishment, is encouraging.

Future Plans

Culturing *N. pectinicornis* will occupy much of our time and resources until field populations have been established at several sites. We will continue with the current release strategy, i.e., inoculating a single waterlettuce infestation several times each week with as many moths (both adults and larvae) as the laboratory colonies at Gainesville and Fort Lauderdale can produce. We will begin inoculating a second site only after we are convinced either that (a) the moth population will persist at the Andytown site or (2) our efforts there are futile. In the latter case, we may choose to further modify the release strategy.

Successful control of several waterlettuce infestations by *N. affinis* clearly demonstrates that responsibility for this weevil should now transfer from research to operations. Consequently, few of our resources will be directed toward *N. affinis* over the coming year. We will, however, survey waterways, as opportunity permits, to monitor dispersal. We will also continue cooperating with interested agencies to increase dispersal of this weevil. This will be accomplished primarily by directing operational personnel to waterlettuce sites with large weevil populations when we discover them and by advising these personnel of the most effective techniques for collection and transport of these biocontrols.



*Figure 2. Sites at which self-perpetuating *Neohydronomus affinis* populations have become established in Florida*

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Further Investigations Into the Effect of Herbivores on Eurasian Watermilfoil (*Myriophyllum spicatum*)

by

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Introduction

Recent investigations of Eurasian watermilfoil declines have found herbivorous insects associated with these declining watermilfoil populations (Painter and McCabe 1988; MacRae, Winchester, and Ring 1990; Creed and Sheldon 1991). The extent to which these herbivores may have contributed to these declines remains to be determined. However, experimental evidence documenting the ability of various aquatic insects to feed on and damage Eurasian watermilfoil in laboratory settings has been accumulating (e.g., Batra 1977; Buckingham and Bennett 1981; Buckingham and Ross 1981; Painter and McCabe 1988; MacRae, Winchester, and Ring 1990; Creed and Sheldon 1991). These observations and experimental results, especially those of recent studies (Painter and McCabe 1988; MacRae, Winchester, and Ring 1990; Creed and Sheldon 1991), suggest that increased research effort needs to be focused on insect herbivory to determine its potential as a control method for nuisance watermilfoil populations.

In Brownington Pond, Vermont, we have found three insect herbivores associated with a watermilfoil population that has undergone a decline (Creed and Sheldon 1991). These insects include two caterpillars, *Acentria nivea* (Lepidoptera: Pyralidae) and *Parapoynx basiusalis* (Lepidoptera: Pyralidae), and a weevil, *Eurhychiopsis lecontei* (Coleoptera: Curculionidae). *Eurhychiopsis* and *Acentria* are much more abundant in Brownington Pond than *Parapoynx* (Creed and Sheldon 1991). Therefore, we have focused our efforts on determining the effects of the first

two herbivores on Eurasian watermilfoil. In a series of laboratory experiments we have already demonstrated that adult weevils (*Eurhychiopsis*) have a strong negative effect on watermilfoil growth (Creed and Sheldon 1991). In this paper, we present the results of two similar experiments documenting the effect of the *Acentria* caterpillars and weevil larvae on watermilfoil growth. In a third experiment we examine the effect of herbivores on watermilfoil buoyancy.

Materials and Methods

Effect of *Acentria* on watermilfoil growth

Several small watermilfoil plants were collected from Brownington Pond. The plants were first checked for herbivore damage. Damaged plants (e.g., with missing meristems, meristem damage, or significant stem damage) were rejected. We selected 18 of the intact plants which were the most similar in size. All obvious invertebrates and weevil eggs were removed from these plants.

The 18 plants were then weighed (blotted wet weight). We then tied a marker around the stem at the base of the plant. The length of the stem from the marker to the tip of the apical meristem was determined. We also counted the number of whorls on each stem above the marker. The initial lengths of the watermilfoil plants ranged from 160 to 203 mm; initial weights ranged from 0.33 to 0.75 g. Much of the variation in weight was attributable to differences in root biomass and not aboveground biomass (Creed, personal observation).

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After processing, each watermilfoil plant was planted in a numbered chamber. Plants were planted in sediment up to the tag on the stem. The chambers were then placed in a large wading pool set out of doors in an unshaded area. The chambers consisted of clear plastic tubes (42-mm inside diameter) set in a polyvinyl chloride pipe base. We first placed aquarium gravel in the bases to weight them. We then filled the remainder of each base with strained pond sediments taken from one of the watermilfoil beds in Brownington Pond. A tight-fitting cap covered with 500- μ Nitex mesh was then placed on the top of the tube. These chambers were of the same type as those described in Creed and Sheldon (1991).

The chambers were aerated with a slow trickle of air bubbles to prevent stagnation. Plants were allowed to acclimate to the chambers for 1 day before the *Acentria* larvae were added.

The experimental design was a randomized complete block design with three treatments per row and six replicates per treatment. The plants were randomly assigned to rows in the wading pool. The determination of treatment within rows was also determined using a random number table. The treatments were 0 (control), 2, or 4 *Acentria* larvae per tube. Two or four *Acentria* larvae were then added to the appropriate chamber in each row. The *Acentria* larvae came from a single batch of eggs that we had collected from Brownington Pond. The larvae had hatched and had been feeding on watermilfoil in an aquarium for about 2 weeks. All larvae used in the experiment were very similar in their initial size (mean length \pm 1 S.E. was 2.8 ± 0.13 mm, based on extra larvae not used in the experiment).

Water temperature in the pool was monitored during the experiment using a max/min thermometer. Water temperatures ranged from 16.1 to 32.2 °C during the experiment (mean minimum temperature was 18.8 °C; mean maximum temperature was 25.8 °C).

The experiment lasted for 22 days. Plants and *Acentria* larvae were then removed from each chamber. After removing the *Acentria*, the watermilfoil plants were measured (from tag to tip of rooted stem) and weighed (blotted wet weight). Any plant material not attached to the rooted stem was not included in the final plant weight. We also counted the number of whorls of leaves remaining on each stem.

Treatment effects were compared using an analysis of variance (ANOVA) with planned, orthogonal contrasts (Sokal and Rohlf 1981). The recovered *Acentria* larvae were preserved in 70 percent ETOH.

Effect of weevil larvae on watermilfoil growth

The design of this experiment, the collection of plants, and the statistical analyses were the same as that discussed above for the *Acentria* experiment. The experiment was conducted in a wading pool adjacent to the one described above. The initial length of the plants ranged from 158 to 223 mm; initial weights ranged from 0.33 to 0.74 g. Late-instar larvae (approximately 3 to 4 mm long) were collected from watermilfoil plants in Brownington Pond the day the experiment was initiated. Treatments consisted of 0 (control), 1, and 2 late-instar larvae per plant. The experiment lasted 9 days. Water temperatures during the experiment ranged from 11 to 27 °C (mean minimum temperature was 15.6 °C; mean maximum temperature was 22.5 °C).

We quantified change in plant length and weight in this experiment. We also measured the amount of stem that had been burrowed by the larvae. As weevil larvae do not appear to feed extensively on leaves, we did not quantify changes in number of leaves. Weevil larvae did not feed on the plants in one replicate from both the 1- and 2-larvae treatments (thus, n = 5 for these two treatments; n = 6 for the control).

Effect of herbivores on watermilfoil buoyancy

In late July we noticed that the tops of watermilfoil plants in the west bed at Brownington Pond had fallen over. The top of the bed was now approximately 1 m below the surface of the pond. Inspection of the plants showed that many of them had been hollowed out by weevil larvae. Many of the collapsed plants showed little or no weevil damage, however, and appeared to have been pulled down by nearby damaged plants. This observation suggested that weevils could have an additional, negative effect on watermilfoil by reducing the buoyancy of the plants. The following experiment was designed to quantify this effect.

We collected undamaged tips of watermilfoil stems and adult weevils on 29 July 1991. The length of the stems was standardized, and they were sorted into 10 groups of six stems each. We then determined the blotted wet weight for each of the groups. The mean wet weight (± 1 S.E.) of the groups of stems was 5.08 ± 0.15 g. The weevils were sexed using a dissecting scope and then sorted into five groups of four weevils (3 females, 1 male). Watermilfoil stems were then placed into ten 38-L aquaria that had previously been filled with well water on 30 July. The aquaria were placed in a line on the ground in an unshaded area on the east side of our research building at Brownington Pond (approximately 15 m from the pond). Each aquarium was aerated with a single airstone. Weevils were added to five of the ten aquaria; the remaining five aquaria served as controls.

Assignment of treatments to aquaria and of watermilfoil and weevils to the aquaria was determined using a random number table. Water temperature was monitored using floating thermometers in three of the control aquaria. Temperatures were recorded in the morning and evening. Temperatures ranged from 16 to 29 °C in the aquaria during the experiment (mean morning temperature was 18.7 °C; mean evening temperature was 23.8 °C).

All aquaria were covered with a tight-fitting lid to prevent the escape of the weevils. The lid consisted of a wooden frame covered with translucent plastic to allow transmission of light. One section of the plastic was removed and replaced with a piece of 500- μ -mesh Nitex that was sealed in place to allow for air exchange and also to help regulate the temperature of the aquaria.

The experiment ran for 21 days. At the end of the experiment, all watermilfoil that was not resting on the bottom was considered to be floating. We should note that all watermilfoil settled to the bottom on cool, overcast days. We sampled the experiment on a sunny day. Floating watermilfoil and that which had settled to the bottom were removed from each aquarium and placed in separate, labeled plastic bags. All herbivores were removed from the aquaria and from the plant material. While this experiment had initially been designed to examine the effects of weevils on buoyancy, we had contamination of four of the weevil tanks with *Acentria* larvae. Therefore, we will refer to the effect on buoyancy as a herbivore effect and not simply a weevil effect.

All watermilfoil was then weighed (blotted wet weight). Most of the watermilfoil in one of the control aquaria had settled to the bottom. The watermilfoil in this aquarium was encrusted with what appeared to be iron precipitation. Using Dixon's test (Sokal and Rohlf 1981) we determined that this replicate was a statistical outlier and removed it from the analysis. Treatment effects were compared using an ANOVA (Sokal and Rohlf 1981). Separate ANOVAs were performed on the weight of floating and sunk plant material. Weight data were log transformed for the ANOVAs.

Results

Effect of *Acentria* on watermilfoil growth

Acentria larvae had a strong effect on change in watermilfoil length (Figure 1a).

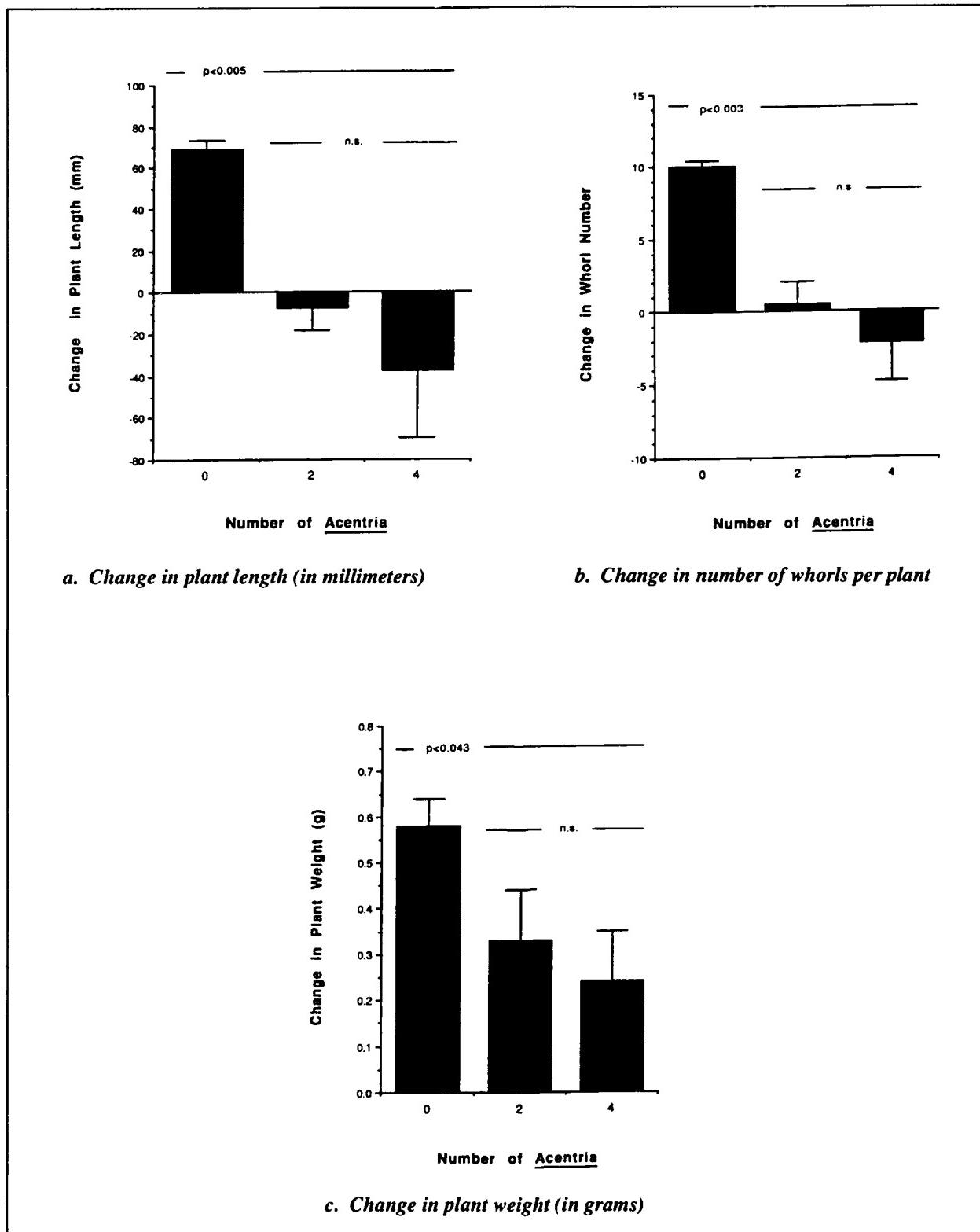


Figure 1. Effect of feeding by *Acentria* larvae on watermilfoil plants. The bars in the histogram represent the mean change in a response variable (± 1 S.E.) for each treatment. The lines with significance values above the histograms show the results of ANOVA comparisons with orthogonal contrasts. In each figure, the upper line represents the comparison of the control versus the *Acentria* treatments; the lower line represents the comparison of the 2- versus the 4-*Acentria* treatment

The mean change in length of the control plants was 68.5 mm compared to -7.5 mm for the 2-*Acentria* treatment and -37.5 mm for the 4-*Acentria* treatment. The contrast between the control versus the two *Acentria* treatments was highly significant. There was no significant difference between the 2- and 4-*Acentria* treatments.

The change in the number of leaf whorls on the stems was similar to the change in length response (Figure 1b). Control plants added an average of 10 new leaf whorls. The contrast between the controls and the *Acentria* treatments was highly significant. Watermilfoil plants with *Acentria* showed either little change in the number of leaf whorls (2-*Acentria* treatment) or a loss of leaf whorls (4-*Acentria* treatment). The two *Acentria* treatments were not significantly different from one another.

Acentria larvae also had a significant effect on change in plant weight (Figure 1c). Control plants gained the most weight and were significantly different from the two weevil treatments. While watermilfoil plants with 2 *Acentria* larvae gained slightly more weight than plants with 4 larvae, the difference was not statistically significant. The observed increase in weight in the two *Acentria* treatments appeared to be almost entirely due to increases in root biomass as length of these plants either did not change or decreased.

Effect of weevil larvae on watermilfoil growth

Weevil larvae did not have a consistent effect on watermilfoil growth (Figures 2a and 2b). The presence of 1 larva reduced watermilfoil change in length compared to the control, but there was no difference in change in length between the 2-larvae treatment and the control. The result of this varied response was that the contrast between the control and the weevil treatments was not significant. However, the contrast between the 1- and 2-larvae treatments was significant

(Figure 2a). Weevil larvae did have a consistent effect on change in weight (Figure 2b). Control plants gained about twice as much weight as either of the weevil treatments. The contrast between the control and the two weevil treatments was marginally significant ($p = 0.08$). The contrast between the two weevil treatments was not significant.

The mean (± 1 S.E.) amount of stem hollowed by weevil larvae in the two treatments was as follows: 1-larva treatment, 75.4 ± 6.7 mm (range 59 to 98 mm); 2-larvae treatment, 106.2 ± 18.6 mm (range 59 to 160 mm). These values translate into burrowing rates of 8.4 mm/day for single larvae and 11.8 mm/day for two larvae. Weevil larvae burrowed through internodes 6 and greater; no burrowing damage was observed in internodes 1-5.

Effect of herbivores on watermilfoil buoyancy

Herbivores had a significant effect on watermilfoil buoyancy. Significantly more watermilfoil was floating in the control aquaria than in the aquaria with herbivores ($F = 19.97$, $p < 0.003$). Significantly more watermilfoil had settled to the bottom in aquaria with herbivores ($F = 205.23$, $p < 0.0001$). Figure 3 shows the buoyancy data plotted as percent watermilfoil floating. Almost all of the watermilfoil (98.6 ± 1.3 percent) was floating in the controls versus only 18.5 ± 7.5 percent for the herbivore treatments.

Not all of the weevil adults were recovered from the tanks at the end of the experiment. Four adults were recovered from two of the tanks, but only two were recovered from the remaining three weevil tanks. Considerable numbers of weevil larvae were produced during the experiment. We removed an average of 26.4 weevil larvae from the five weevil tanks (range 11 to 43). Three dead weevil pupae and one *Acentria* larva were found in the control aquaria. The numbers of *Acentria* found in the weevil tanks were 0, 1, 2, 3, and 18.

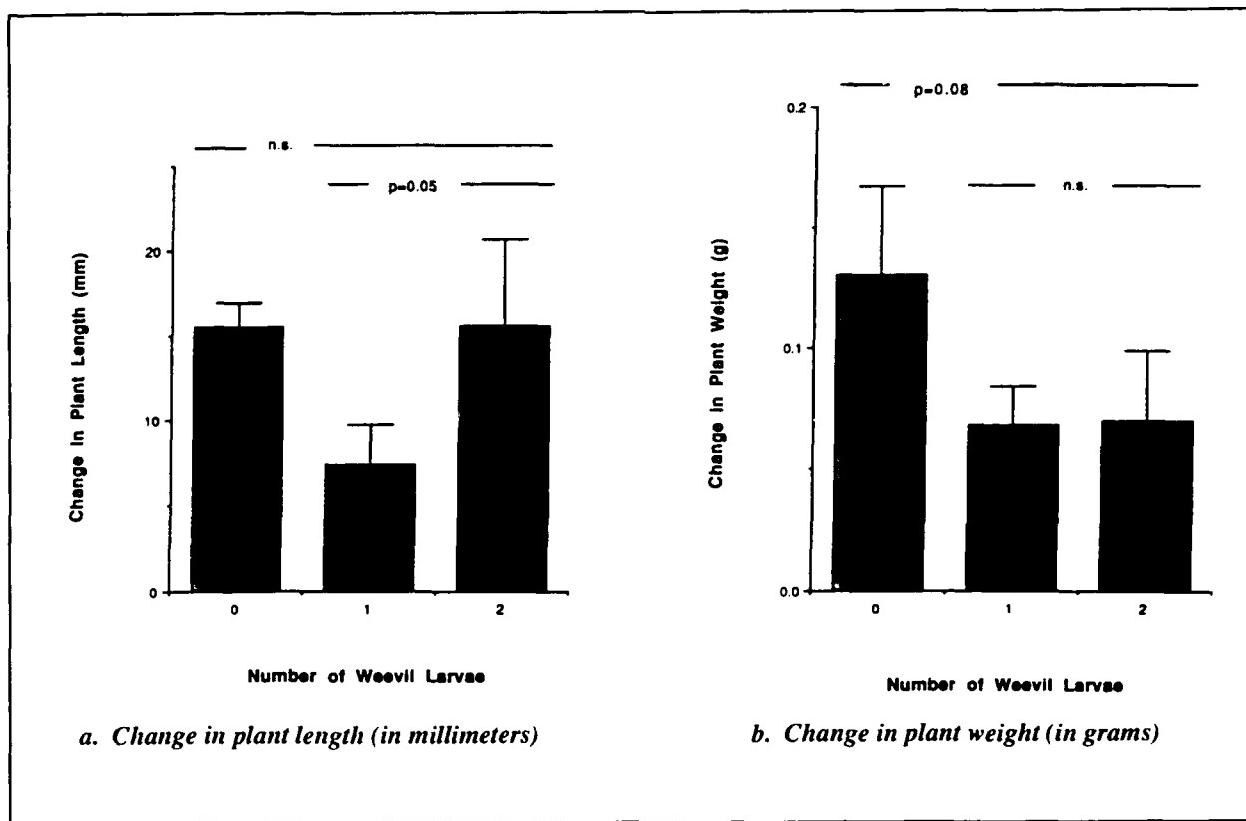


Figure 2. Effect of feeding by *Eurhychiopsis* larvae on watermilfoil plants. The bars in the histogram represent the mean change in a response variable (± 1 S.E.) for each treatment. The lines with significance values above the histograms show the results of ANOVA comparisons with orthogonal contrasts. In each figure, the upper line represents the comparison of the control versus the weevil treatments; the lower line represents the comparison of the 1- versus the 2-weevil treatment

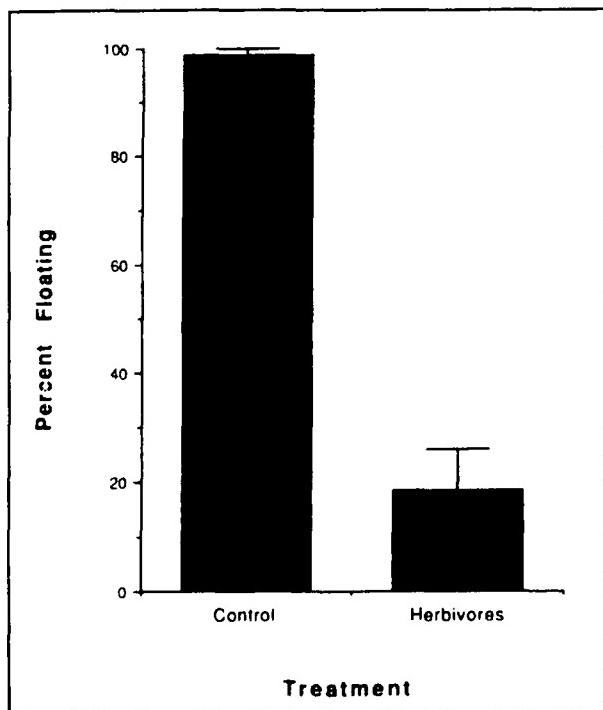


Figure 3. Effect of herbivores on watermilfoil buoyancy. The bars in the histogram represent the mean (± 1 S.E.) percent of total watermilfoil weight found floating in each of the two treatments

Discussion

The larvae of *Acentria* and *Eurhychiopsis* had somewhat different effects on watermilfoil. *Acentria* had a pronounced effect on both measured components of watermilfoil growth, i.e., change in length and weight. The effect of *Acentria* appears to differ from that of the *Eurhychiopsis* larvae as a result of the different feeding modes of these animals. *Acentria* feed largely on the exterior of the plant. While first instars of *Acentria* have been reported to burrow into the stem (Batra 1977), most of the larval phase is spent on the outside of the plant feeding on leaf material. (Note: We have observed late-instar *Acentria* larvae inside of watermilfoil stems collected from beneath the ice in Brownington Pond.)

The effect on length and weight is largely a result of larger *Acentria* larvae cutting the stem to build their retreats. However, larvae may cut the stem additional times. We noticed that two of the plants with four larvae had been cut into more pieces than would be expected for retreat construction. This also occurred in the tank in the buoyancy experiment that had 18 *Acentria*. The plants were cut into numerous small pieces that had settled to the bottom.

The effect of *Eurhychiopsis* larvae on watermilfoil length and weight was not as strong as that of *Acentria*. This is due, in part, to the fact that we used late-instar *Eurhychiopsis* larvae in this experiment. While first-instar weevil larvae feed on apical tissue and thus directly affect apical growth, late-instar larvae feed by burrowing through the stem. As much of the plant weight is in stem tissue, removal of substantial portions of stem tissue should have an impact on weight. Late-instar larvae do not feed on meristematic tissue and thus should not be expected to have a direct effect on length change. However, removal of stem vascular tissue could indirectly influence stem elongation as a result of reduced or halted translocation of nutrients from roots to actively growing portions of shoots. If adequate quantities of nutrients

can be removed directly from the water by the plant, this effect should be negligible.

While the sediments are an important source of nutrients for rooted aquatic macrophytes (e.g., Barko and Smart 1978, 1981), *M. spicatum* can absorb nutrients from the water column through stem and leaf tissue (Best and Mantai 1978). The ability of *M. spicatum* to take up water column nutrients appears to be a function of nutrient concentration (Best and Mantai 1978), with nutrients being absorbed from the water when at higher concentrations. Thus, larval burrowing might have a more pronounced effect on *M. spicatum* growth in nutrient-poor water bodies if the growing portion of the stems cannot obtain sediment nutrients.

While the above information suggests that feeding by only late-instar weevil larvae on watermilfoil could produce a variable response in length change, it does not account for the consistent difference between the 1- and 2-larvae treatments observed in our experiment for this response variable. We are unable to account for this effect.

One further comparison needs to be made between the effect of *Acentria* and *Eurhychiopsis* larvae on watermilfoil. Larval weevil burrowing weakens the watermilfoil stem, with the result that burrowed stems are easily broken. While we have commonly encountered such broken stems in lakes and ponds, this effect was not observed in our experiment as the stems were protected from physical disturbance by the chambers.

Herbivores clearly had a strong effect on watermilfoil buoyancy. This result is not surprising as both *Acentria* and *Eurhychiopsis* (both adults and larvae) expose stem vascular tissue while feeding. We have observed adult and larval weevils feeding on several occasions, and it is not unusual to see a stream of bubbles emerging from the damaged portion of the stem. That this small-scale effect could result in the collapse of the upper portion of a watermilfoil bed is significant. The implication is that this suite of herbivores

does not have to remove considerable amounts of stem and leaf tissue to have a strong negative effect on watermilfoil. The consequences of leaf removal may be minor in comparison to the effect of loss of buoyancy. If herbivore feeding can cause plants to drop out of the photic zone (or at least well-lit surface waters), the plants may be unable to recover from this damage. The effect becomes even more pronounced if damaged plants can drag down healthy ones. Indeed, loss of buoyancy may prove to be one of the major mechanisms of watermilfoil bed destruction by herbivores.

To date, three studies have found herbivorous insects associated with declining watermilfoil populations (Painter and McCabe 1988; MacRae, Winchester, and Ring 1990; Creed and Sheldon 1991). In all three instances, the researchers conducted laboratory experiments that demonstrated that the associated herbivores could have negative effects on watermilfoil.

Painter and McCabe (1988) found that watermilfoil could tolerate and continue to grow with low densities of *Acentria*. At higher densities, *Acentria* had a negative effect of watermilfoil growth. However, Painter and McCabe did not determine how *Acentria* feeding could generate these results.

MacRae, Winchester, and Ring (1990) conducted more detailed experiments that evaluated the effects of a midge larva (*Cricotopus myriophylli*) on watermilfoil growth. They found that this midge, which feeds on just the meristems, could prevent the plants from increasing in length or weight. Only one larva per apical tip was needed to produce this effect.

We have obtained similar results in an experiment that examined the effects of adult weevils (*E. lecontei*) on watermilfoil (see Experiment 1, Creed and Sheldon 1991). Adult weevils can influence elongation by destroying

meristems. In that experiment, some of the effect on change in length was attributable to first-instar larvae, which also feed on the meristem (see Creed and Sheldon 1991, for details). Adult weevils had an even stronger effect on change in weight. At densities of four weevils per plant, watermilfoil plants lost weight during the experiment. This resulted primarily from the removal of several leaves.

All of these studies have focused on the impact of these herbivores on watermilfoil growth. However, our field observations and the buoyancy experiment suggest that herbivores (at least weevils) may do more than just suppress growth. They may have an additional negative impact on watermilfoil plants if they can cause them to sink.

The results discussed above, along with those presented in this paper, suggest that herbivores might be playing an important role in either reducing the abundance of watermilfoil or maintaining populations at manageable levels. While it remains to be demonstrated that any of these herbivores can cause a decline, the data collected from laboratory experiments are promising. Continued effort should be focused on determining the mechanisms by which these herbivores affect watermilfoil. However, we are rapidly reaching the stage where we need to conduct controlled herbivore introductions to fully assess the ability of these insects to suppress or reduce nuisance watermilfoil growth.

Acknowledgments

We wish to thank Kristin Henshaw, Diana Cheek, and Gabe Gries for their invaluable help in conducting these experiments. This work was funded by the US Environmental Protection Agency Clean Lakes Demonstration Program, the US Army Corps of Engineers (Contract No. DACW39-90-K-0028), the Vermont Department of Environmental Conservation, and Middlebury College.

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Biological Control of Aquatic Weeds Using Plant Pathogens

by

Judy F. Shearer¹

Introduction

The plant pathogenic fungus *Mycoleptodiscus terrestris* (Gerdemann) Ostazeski (Mt) has been shown in previous greenhouse studies to significantly reduce plant biomass of *Myriophyllum spicatum* L., Eurasian water-milfoil (Gunner et al. 1990, Stack 1990, Smith and Winfield 1991). *Mycoleptodiscus terrestris* can exhibit growth over a wide temperature range (10 to 35 °C) (Gerdemann 1954) but grows optimally between 20 and 30 °C.

The purpose of the study described in this paper was to determine the range of temperatures at which formulated Mt (Aqua-Fyte) (EcoScience Corporation, Worcester, MA) can impact Eurasian watermilfoil resulting in the greatest reduction in plant biomass. The information will then be used to optimize application times for Aqua-Fyte release in field trials.

Materials and Methods

Clear acrylic tubes 1.5 m long and 13.75 cm in diameter were used for growth chamber studies. Lake sediment amended with NH₃Cl was placed in the bottom of each tube and covered with 7.5 cm of washed silica sand. Three 15-cm sprigs of fresh *M. spicatum* were planted in each tube, and 18 L of nutrient solution (Smart and Barko 1985) was added. The tubes were aerated and maintained in temperature-controlled tanks at 23 ± 1 °C under natural light conditions in a greenhouse. When plant height reached approximately three-quarters the height of the tube, the tubes were

placed in a growth chamber set on a 14/10-hr light/dark cycle. The plants were allowed to acclimate to the desired experimental temperature of 15, 20, or 25 °C for 7 days.

Treatments were arranged in the growth chamber in a randomized block design with five replications. The experiments were performed serially with each experiment being conducted twice.

Mt-treated plants were inoculated with 16.0 g of Aqua-Fyte in the form of alginate/clay pellets containing the pathogen. Controls consisted of either untreated plants or plants treated with 16.0 g of a formulation control of alginate/clay pellets minus the active ingredient Mt. Four weeks after inoculation, the remaining living biomass in each tube was collected, separated into roots and shoots, dried at 80 °C, and weighed. Three 2-cm pieces of stem tissue were collected respectively from basal, median, and apical sections of plants in each tube for microbial analysis. Pellets were retrieved from inoculated tubes for viability determinations following submersion for 4 weeks.

Harvested stem segments were surface sterilized in a 1.5-percent hypochlorite solution for 15 sec, rinsed in sterile distilled water, and blotted dry on sterile paper towels. Stem pieces and harvested pellets were plated on Martin's agar (Martin 1950) and incubated at 28 °C for 4 days. Presence of Mt was confirmed by visually examining the plates for characteristic fungal colony growth. Percent infection was based on the number of stem pieces or pellets that developed Mt colonies.

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Results

Four weeks after inoculation, Aqua-Fyte significantly reduced the aboveground biomass of milfoil at 20 and 25 °C compared with the formulation control or untreated plants (Figure 1). The reduction in aboveground biomass ranged from 73 to 75 percent between controls and Aqua-Fyte-treated plants at 25 and 20 °C. No significant reduction in aboveground biomass between treated and control plants occurred at 15 °C.

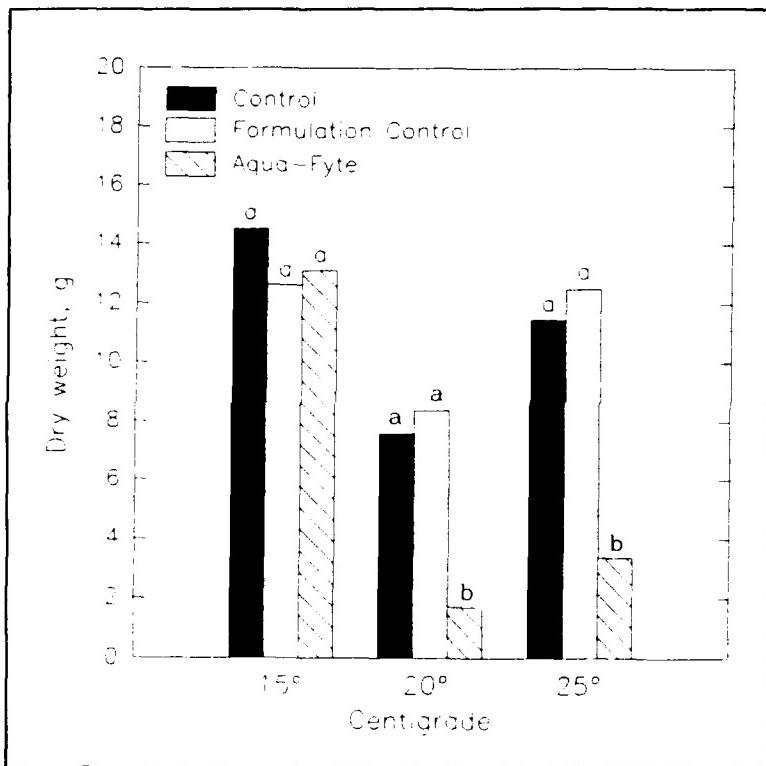


Figure 1. Aboveground biomass of treated and untreated Eurasian watermilfoil plants grown for 4 weeks at 15, 20, or 25 °C

There were no significant differences in the belowground biomass of Aqua-Fyte-treated plants and the controls at 15 and 20 °C (Figure 2). At 25 °C, belowground biomass of Aqua-Fyte-treated Eurasian watermilfoil plants was reduced by 59 percent when compared with the untreated control plants.

Root-shoot ratios were not significantly different between the Aqua-Fyte-treated plants and the controls at 15 °C (Figure 3).

At 20 and 25 °C, plants inoculated with Aqua-Fyte had substantially higher root-shoot ratios than either the uninoculated controls or the formulation controls. High ratios of root-to-shoot biomass have been documented to occur when plants are grown in infertile environments (Chapin 1980). Stress not directly related to plant nutrition has also been suggested as an additional contributing factor to high root-shoot ratios (Barko 1983).

Mt was not recovered from stem sections or pellets of formulation control-treated plants or from stem sections of untreated control plants. Mt was consistently recovered from basal and median stem sections of Aqua-Fyte-inoculated plants at 15, 20, and 25 °C (Figure 4). At 15 °C, no colonies of Mt grew from apical stem pieces, but at 20 and 25 °C, 30 and 14 percent, respectively, of the apical pieces produced characteristic Mt colonies on Martin's agar. Reisolation of Mt colonies from recovered Aqua-Fyte pellets was 100, 90, and 88 percent at 15, 20, and 25 °C, respectively.

Discussion

In growth chamber studies, Aqua-Fyte was shown to substantially reduce aboveground biomass of Eurasian watermilfoil at 20 and 25 °C but not at 15 °C. These data would suggest that water temperatures under natural field conditions must reach at least 20 °C

for the strain of Mt tested in the study to be effective in controlling Eurasian watermilfoil. The upper temperature limit at which Mt substantially reduces biomass has yet to be determined, but fungal growth curves suggest that it is less than 30 °C and probably around 28 °C. Because Mt requires a period of attachment to a host species before primary invasion of the fungus can occur, the Aqua-Fyte pellets must come into direct contact with milfoil plant tissues for the inoculum to be

Figure 2. Belowground biomass of treated and untreated Eurasian watermilfoil plants grown for 4 weeks at 15, 20, or 25 °C

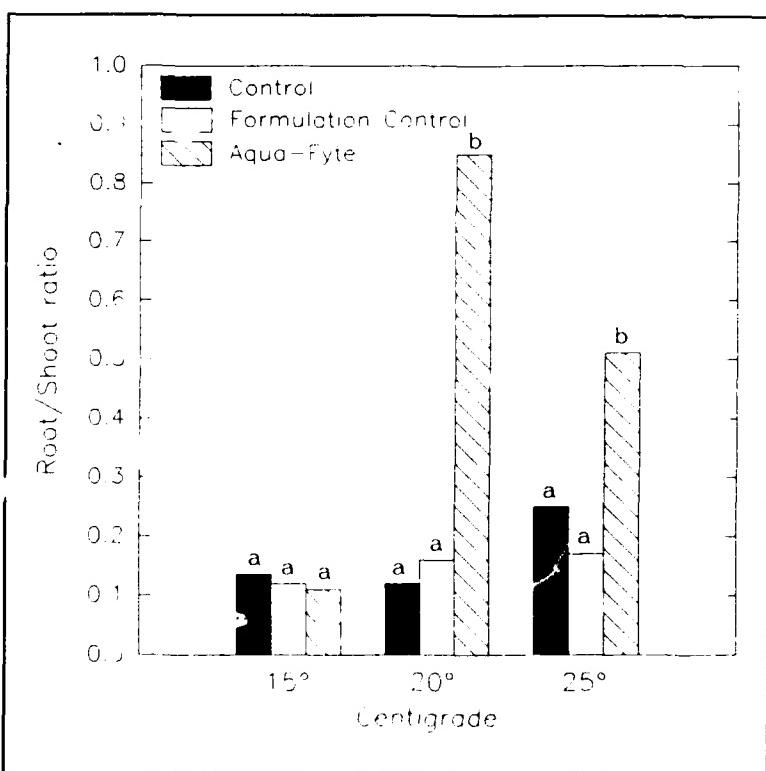
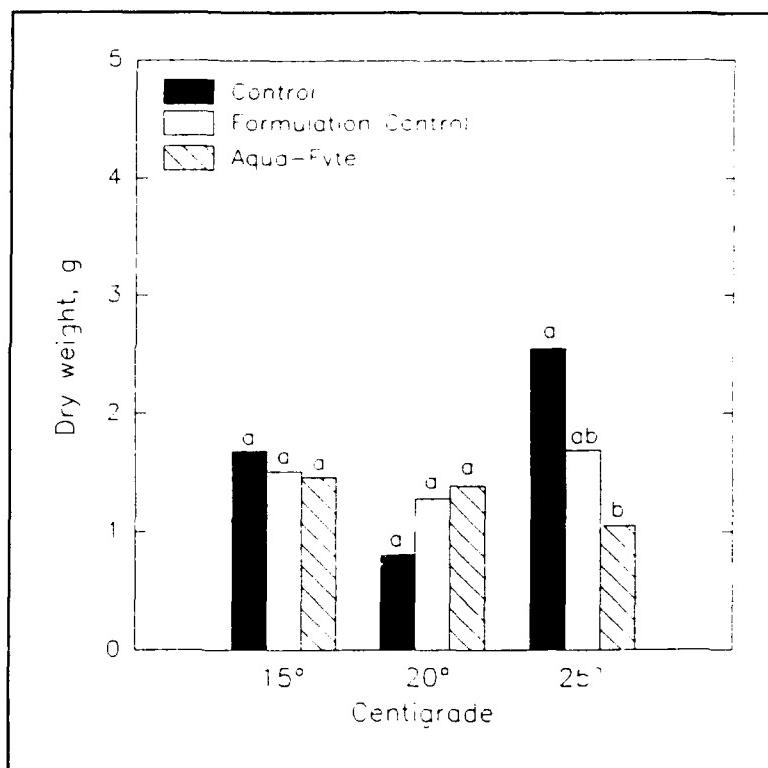


Figure 3. Ratio of root-to-shoot biomass of treated and untreated Eurasian watermilfoil plants grown for 4 weeks at 15, 20, or 25 °C

effective. Differences in the percent recovery of Mt from plant tissues is most likely a function of site of attachment of the primary inoculum from the Aqua-Fyte pellets and site of invasion by secondary inoculum produced by the fungus during the experiment. Since the pellets fall through the water column in the tubes, higher recovery rates would be expected from basal and median stem pieces than from the apical regions of inoculated plants.

Although Mt was able to invade basal and median stem sections at 15 °C, fungal growth rate was so slow that a substantial reduction in biomass did not occur. However, because Mt was recovered with high frequency from the pellets at low temperatures, it is possible that a field application could be undertaken at low temperatures and still be effective when water temperatures approach and exceed 20 °C, provided the pellets remain in close association with the plant over time. Optimally, to have the greatest impact on Eurasian watermilfoil, Aqua-Fyte should be applied when water temperatures are between 20 and 25 °C.

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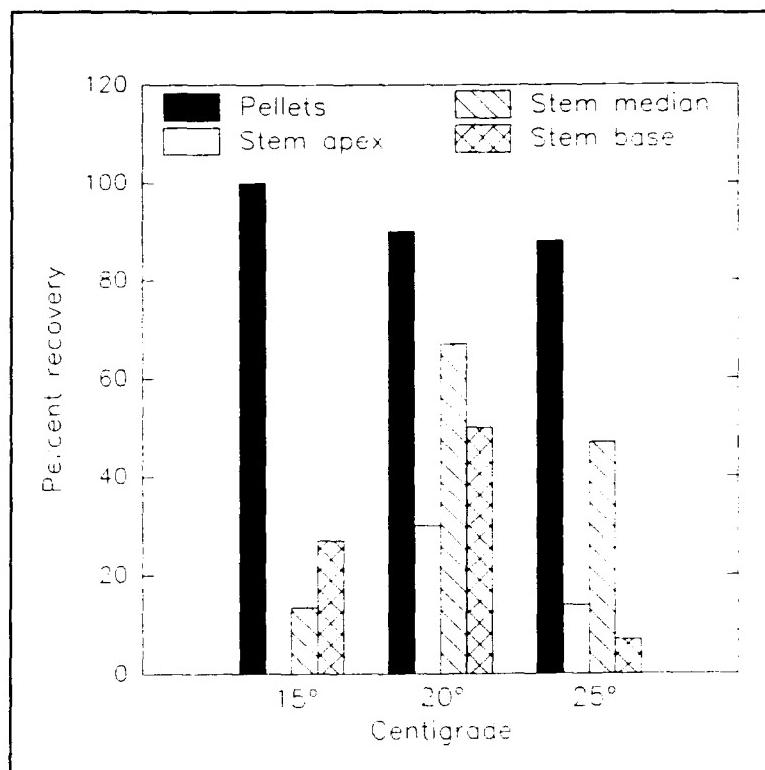


Figure 4. Percent recovery of Mt from pellets and basal, median, and apical stem pieces of Aqua-Fyte-inoculated Eurasian watermilfoil plants grown for 4 weeks at 15, 20, or 25 °C

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The Allelopathic Ability of Three Species of Aquatic Plants To Inhibit the Growth of *Hydrilla verticillata* (L.f.) Royle

by
Harvey L. Jones¹

Introduction

Background

Hydrilla verticillata (L.fil.) Royle (common name, hydrilla) is a noxious aquatic plant introduced into the United States from Africa through the aquarium industry and sold under the name "oxygen plant" or "star vine." Hydrilla has long branching stems that often fragment and form large floating mats (Tarver et al. 1980), and can grow in water depths up to 15 m.

Two reproductive structures enable hydrilla to withstand extremely harsh weather conditions: (a) turions or winter buds (dense clusters of apical leaves that are produced in the leaf axils, green and ovoid-conical shaped buds) and (b) bulb-like hibernacula, commonly but incorrectly called tubers (which are formed at the ends of stolons buried in the substratum).

Hydrilla plants are found in lakes, rivers, drainage and irrigation canals, ponds, and streams. Severe infestations of hydrilla can restrict boat traffic and interfere with fisheries and water flow.

Hydrilla is a nuisance adventive submersed aquatic plant that reproduces by fragmentation, by tubers (which may remain dormant in the substrate for several years and yet remain viable), by turions, and by seeds (in the monoecious variety). This plant is one of the most prolific nuisance aquatic plants in the United States, causing problems in many lakes and reservoirs with recreation and navigation.

Hydrilla is found in many southern states and California, and recently Virginia.

Hydrilla is extremely difficult to control because of its varied methods of reproduction. Mechanical removal methods tend to increase the spread of hydrilla, as a result of fragmentation, and while herbicides are used in numerous places, there is major concern for the environment and water quality.

Allelopathy refers to the biochemical interactions that take place among plants; however, its effectiveness depends on the addition of a chemical to the environment (Sutton 1986a). In general, the term allelopathy refers to the detrimental effects of higher plants of one species (the donor) on the germination, growth, or development of another species (the recipient) (Putnam 1985).

Rice (1974) provided us with a more functional definition of allelopathy as being any direct or indirect harmful effect by one plant (including microorganisms) on another through production of chemical compounds that escape into the environment. Similarly, Parker (1984) defined allelopathy as the harmful effect of one plant or microorganism on another owing to the release of secondary metabolic products into the environment. Allelopathy may be a potentially important mechanism in the management of undesirable aquatic plants. Because it may provide an inexpensive and more desirable method of control than more conventional methods such as the use of herbicides or mechanical removal, allelopathy may prove to be one of our best methods to control hydrilla.

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Studies have shown that some plants have the ability of controlling the distribution, in some cases completely eradicating other species from that same area. Most of these studies, however, have been conducted using terrestrial plants or plants located in the littoral zone of aquatic habitats. Few studies have shown the allelopathic effects of submersed aquatic hydrophytes.

The purpose of this research was to conduct experiments with rooted plants, to which plant organic matter had been added to the substrate, to determine if results similar to those found during previous test tube assays using plant extracts could be obtained.

The specific objective of this research was to test the three aquatic plant species for their ability to reduce the growth, reproduction, and/or distribution of the hydrophyte hydrilla. Procedures used in this study were modified from Barko and Smart (1983).

Test species selection

Species selected for analysis during this study were based on test tube assays with 15 plant extracts conducted in the laboratory at the Waterways Experiment Station (WES) (Jones and Kees 1991). From these studies we found that three species, coontail (*Ceratophyllum demersum*), eelgrass (*Vallisneria americana*), and pondweed (*Potamogeton nodosus*), were potentially allelopathic to hydrilla. The three species that appeared to be allelopathic in the test tube assays were then used in the rooted plant study.

Test tube assays were used first because this is a rapid screening method to indicate the allelopathic potential of various candidates. The next logical step in the research effort was to conduct rooted plant studies (tank studies), which represent the next scale up from the test tube assays.

Through laboratory bioassays, Elakovich and Wooten (1989) listed several species that were potentially allelopathic to hydrilla. The plant species used in this test were selected because they were the three best candidates

identified as potentially allelopathic to hydrilla as a result of tube assays previously conducted in greenhouse experiments (Smith and Jones 1990, Jones and Kees 1991).

Methods and Materials

Plant collection

Aquatic plants were field collected from Caddo Lake, Louisiana, and from J. D. Murphree Wildlife Refuge, Port Arthur, Texas, and transported to Vicksburg, MS, in ice chests with sufficient ice to prevent the plants from deteriorating. Entire plants were collected whenever possible. Plants were washed to remove dirt and debris, oven-dried at 70 °C, and ground with a Wiley mill to 7 µm.

Hydrilla stock cultures

Stock cultures of hydrilla were grown from field-collected specimens and maintained in the greenhouse in 1,150-L fiberglass tanks. Plants were aerated with compressed air and maintained at a constant temperature of 25 °C. Plants were grown in lake sediment and recultured periodically using apical tips.

Experiment 1. In a greenhouse test, 5 and 20 percent organic matter from *C. demersum*, *P. nodosus*, and *V. americana* were added to 32-oz. plastic cups and 1.248 g of lake sediment (Brown's Lake, MS) and mixed thoroughly. Three 15-cm-long apical tips of hydrilla were placed in the plastic cups to a depth of 10 cm, then overlain with a layer of silica sand to prevent sediment from leaching into the water column. The cups were placed in 30-in.-tall (12-L capacity) Plexiglas cylinders, then filled with nutrient solution (modified Barko's medium). The acrylic columns were then placed in 1,150-L capacity fiberglass tanks filled with tap water (as a water bath) and maintained at a constant temperature of 25 °C.

Aeration was supplied to each column through the use of compressed air, to prevent algal growth and ensure proper mixing of chemicals in the nutrient solution. The study was allowed to run for a period of 4 weeks.

and was then repeated two additional times for verification of results.

Data analyses. The data were subjected to analyses of variance (ANOVA) and the Duncan's Multiple Range Test of the Statistical Analysis System (SAS) to determine if significant differences existed between the controls and test species.

Results and Discussion

According to the analyses of data from all three experiments, there were no significant differences between experiments. Therefore, data from all three experiments were combined and analyzed using the ANOVA of SAS. Test results showed no significant differences in total biomass with 5-percent concentration of organic matter addition; however, with the 20-percent concentrations, *Ceratophyllum* showed significant differences in biomass (Figure 1) when compared to control.

When mean lengths for the two concentrations of matter additions were compared at the 5-percent concentration, no significant differences were detected; however, at the 20-percent concentration, *Ceratophyllum* and *Vallisneria* were found to be significantly different from the control (Figure 2), while borderline significant differences were detected for *Potamogeton*.

Conclusions

Based on the results of this study, we have found *Ceratophyllum* to be our best candidate for being allelopathic to hydrilla. A couple of other species are also worth additional attention because they were borderline allelopathic in various phases of our study. These species include *Vallisneria* and *Eleocharis*, since it has been found to be allelopathic to several aquatic plant species. When grown in established stands of spikerush, hydrilla biomass production was decreased by 85 to 90 percent when compared to hydrilla plants grown alone (Sutton 1986b).

Future Work Planned

This investigation reinforces the results of earlier studies conducted at the WES (Jones and Kees 1991), which have indicated that *Ceratophyllum* appears to be the best potential allelopathic candidate to impact the growth of hydrilla. We will be conducting field tests with hydrilla and the various types of organic matter found to be allelopathic in this study. In addition to *Ceratophyllum*, we will also look at *Vallisneria* and *Eleocharis* more closely, since they were found to be allelopathic to hydrilla in various parts of our study.

Future research will include conducting test tube assays using aquatic plant extracts and rooted plant studies (tank) with Eurasian watermilfoil as the target species, with macerated plant material identified through the test tube assays as being the most allelopathic to milfoil.

Acknowledgments

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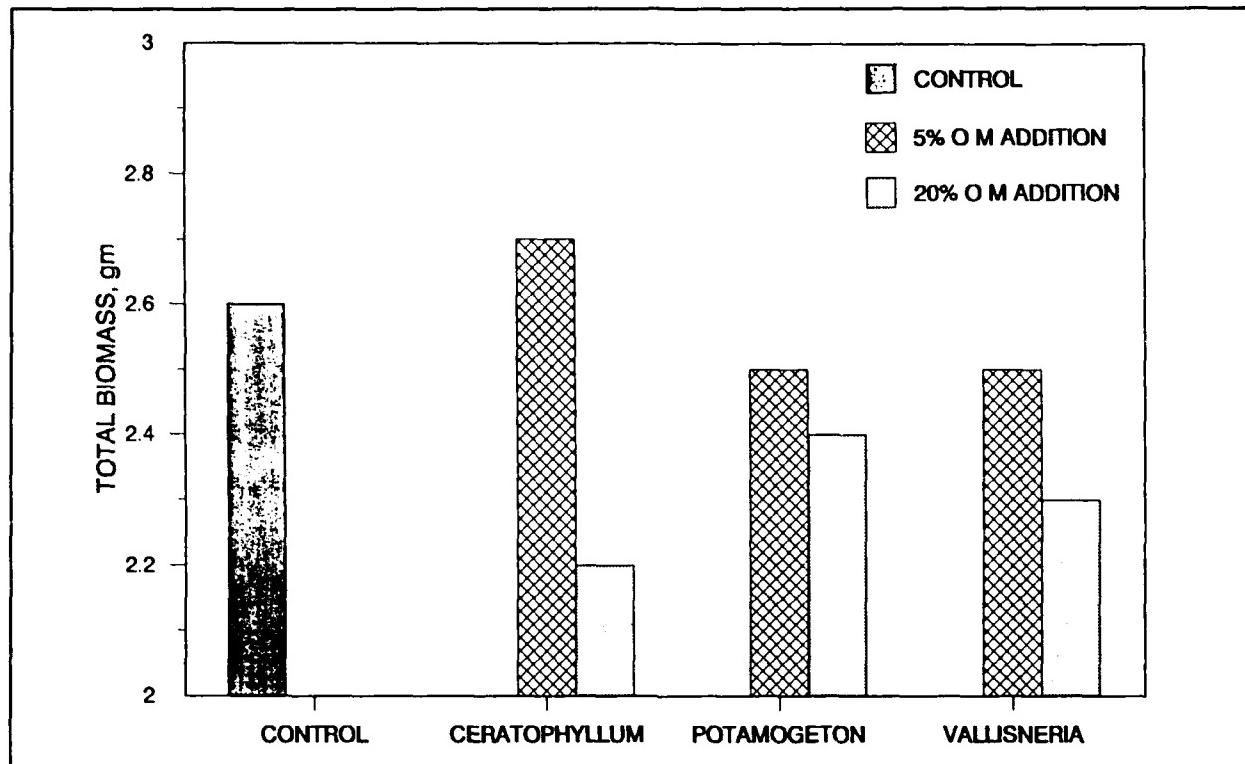


Figure 1. Allelopathic influence of organic matter additions (5 or 20 percent dry weight) on aboveground biomass of *Hydrilla verticillata*. Values are the means of three replicates

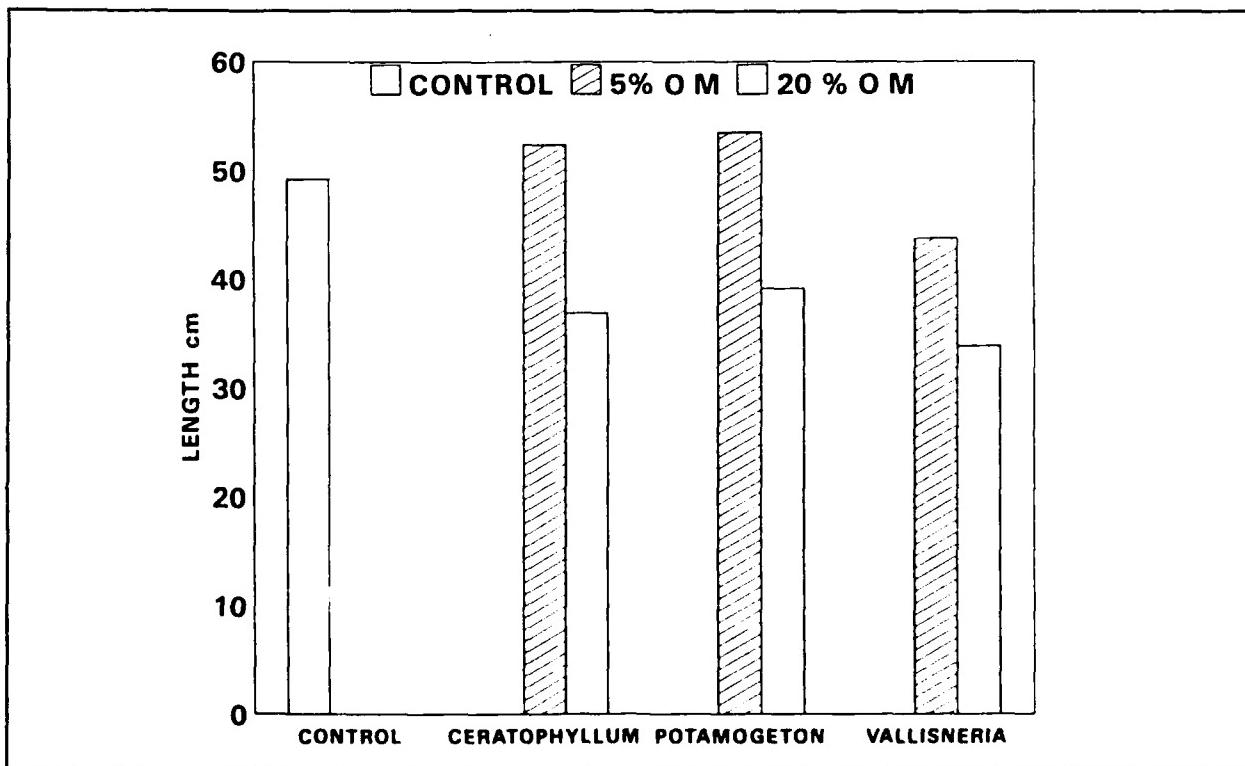


Figure 2. Allelopathic influence of organic matter additions (5 or 20 percent dry weight) on the mean length of *Hydrilla verticillata*. Values are the means of three replicates

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